Local Solvation Behavior in Micro-heterogeneous and in Phase-separated Aqueous Alcohol Solutions

Subhadip Chakraborty [1] , **Debasish Das Mahanta** [2,*]

[1]   Centre for Healthcare Science and Technology, Indian Institute of Engineering Science and Technology, Shibpur, Howrah-711103, India
[2]   Department of Physics, Gandhi Institute of Technology and Management (GITAM) University, Bengaluru, Karnataka, 561203, India
*   Correspondence: snbdebasish@gmail.com

Abstract: This research paper delves into the ongoing discourse about the correlation in the structural and dynamic attributes of hydrogen bond (HB) networks within alcohol-water binary mixtures. Employing Terahertz time-domain spectroscopy (TTDS) and Terahertz-Fourier transform infrared (THz-FTIR) spectroscopy, we conduct an in-depth exploration of the intermolecular roto-vibrational modes of water. Through this analysis, we extract relevant insights concerning the configuration of the HB network. Our study reveals a comprehensive comprehension of the variances in structure and dynamics within the HB network in response to alterations in solute concentration. We have considered both micro-scale and macro-scale for partially water-soluble alcohol phases. Furthermore, we have considered the influence of hydrophobicity and the macroscale self-aggregation tendencies of alcohols, culminating in a holistic viewpoint of the intermolecular networks within these binary mixtures. We found that the delicate equilibrium between local homo-molecular structures with the cross-molecular solvation configurations leads to micro/macro scale self-aggregation that exerts significant influence over the collective behavior of the mixture.

Keywords: hydrogen bond structure and dynamics; bio-condensate; phase separation; Terahertz spectroscopy; self-associated clusters; intermolecular interactions.

1. Introduction

Water, an omnipresent and crucial solvent, exerts significant sway over the arrangement and operations of dissolved particles due to its intermolecular hydrogen bond (HB) [1-2]. Despite extensive dedication to investigating the molecular arrangements and dynamic reactions of substances within the aqueous medium, exploring this field remains an essential realm of inquiry [1]. Alcohol-water mixtures are pivotal in various biological, chemical, and engineering scenarios and have been the subject of thorough examination owing to their intriguing physicochemical characteristics [3]. Alcohol molecules have captivated considerable research attention due to their ability to extensively modulate physicochemical attributes by selecting optimal alkyl chain lengths or exploiting diverse potential isomeric configurations [4]. These molecules are inherently amphiphilic, encompassing both hydrophilic and hydrophobic segments, and as a result, they elicit contrasting effects on water, a phenomenon often termed '*The Janus Effect*' [5]. The hydrophilic constituents have a robust propensity to engage with water by forging HBs, while the hydrophobic components tend to amass into aggregates via self-association, thereby prompting perturbations in the water's

structure - an occurrence commonly recognized as '*hydrophobic hydration*' [6]. A multitude of experimental methodologies has been employed to scrutinize aqueous alcohol solutions, encompassing Raman spectroscopy [7], neutron diffraction [8], nuclear magnetic resonance (NMR) [9], infrared spectroscopy [10-11], mass spectrometry [12], dielectric spectroscopy [11, 13-15], etc.

Additionally, numerous numerical and computational inquiries have contributed to a holistic comprehension of the attributes exhibited by these solutions from a molecular perspective [11, 16-18]. Despite a strenuous endeavor, establishing a definitive correlation between these systems' HBonded structure and dynamics remains elusive, given that measurements frequently yield seemingly contradictory outcomes. This circumstance underscores the necessity to delve into alterations in the kinetics of such blends as they adapt their microstructures based on the composition of solutes.

Terahertz (THz) spectroscopy emerges as a powerful technique for investigating molecular reorientation and related intermolecular vibrations - referring to the localized fluctuations of dipoles - coexisting within this frequency range, thus contributing to the overall dynamics of liquids [19-24]. These intermolecular movements are primarily influenced by the HB network. Past studies utilizing dielectric relaxation (DR) measurements on alcohol-water mixtures within the gigahertz (GHz) frequency range faced limitations in time scales, hindering precise observation of the cooperative relaxation of water molecules around small solutes [25]. In contrast, DR measurements within the THz frequency realm offer an accurate perspective on the picosecond (ps) to sub-ps collective behaviors of the HB network, spanning multiple hydration layers around solute surfaces - a phenomenon often obscured in conventional spectroscopic methods [14, 26-27].

An investigation by Schmuttenmaer *et al*. demonstrated that straightforward DR models could aptly elucidate experimentally derived intricate dielectric constants for water and single alcohols, extending up to at least 1 THz (10^{12} Hz) [28]. This elevated dielectric parameters to a pivotal role as benchmarks for molecular dynamics (MD) simulations. Notably, this research excluded the presence of mixed alcohols in water. In separate reports, Yomogida *et al*. examined temperature-dependent dielectric attributes of monohydric alcohols using TTDS. The expansive vibration mode displayed temperature independence and was ascribed to the oscillation of alkyl groups within the interconnected HBonded chain-like arrangement. However, existing literature lacks coverage of high-frequency modes and their associated dynamics [29]. Sarkar *et al*. adopted a combined broadband TTDS approach spanning from 0.5-10 THz, coupled with simulations, to portray the dielectric characteristics of methanol, ethanol, 1-propanol, 2-propanol, and 1-butanol [13]. However, this study primarily pertains to alcohol in bulk and does not encompass the intermolecular interactions between the constituent molecules present within binary mixtures. In recent times, we have utilized both experimental (THz-TDS and THz-FTIR spectroscopy) as well as classical MD simulations to investigate the shifts in local solvation structures during the occurrence of liquid-liquid phase separation driven by solute aggregation within aqueous binary mixtures [19, 30-31].

In this present research, we've delved into the THz dielectric response exhibited by water-alcohol binary mixtures, connecting this response to the structure and kinetics of the HB network, especially in the context of the distinct phase behavior displayed by these solutions. Our study encompasses a range of monohydric alcohols with varying hydrophobic chain lengths and alcohols with diverse isomeric configurations and solubility characteristics. These isomer variations result in distinct phase behaviors when these alcohols are introduced into

aqueous solutions. For instance, methanol (MOH) and tert-butanol (TBA) are fully soluble in water. Conversely, 1-butanol (1BA) is only partially soluble in water. **Scheme 1** provides an overview of the chemical structures of the alcohol molecules utilized in our study and highlights the differing phase behaviors between fully soluble and partially water-soluble alcohol mixtures. Notably, even with the same chemical formula, 1BA and TBA exhibit distinct phase behaviors in their aqueous mixtures. TBA, for instance, is miscible in water across all concentrations, whereas 1BA is only partially soluble in water under typical conditions, leading to observable macroscopic phase separation due to the formation of microscopic self-associated clusters [19, 30, 32].

Our investigation employs both THz-TDS and THz-FTIR spectroscopy, spanning a broad frequency range from 0.3-18 THz. This extensive range captures various intermolecular roto-vibrational modes within the HB-network of water in alcohol-water binary mixtures. It effectively captures the rotational dynamics of the solutions, offering explicit insights into the intermolecular translational and librational motions of HBs as water interacts with alcohol molecules. Through this study, we've outlined the structure and kinetics of the HB network within alcohol-water binary mixtures, examining how it's influenced by the composition of solutes, taking into account both the microscale (for fully soluble alcohols) and macroscale (for partially water-soluble alcohols) phase behaviors. Additionally, our research encompasses the effects of both hydrophobic chain lengths and macroscopic self-aggregation of alcohols, which, individually or collectively, can perturb the structure of water. This comprehensive investigation offers a collective perspective on the interplay between HB-networks of alcohol-alcohol, alcohol-water, and water-water within these binary mixtures.
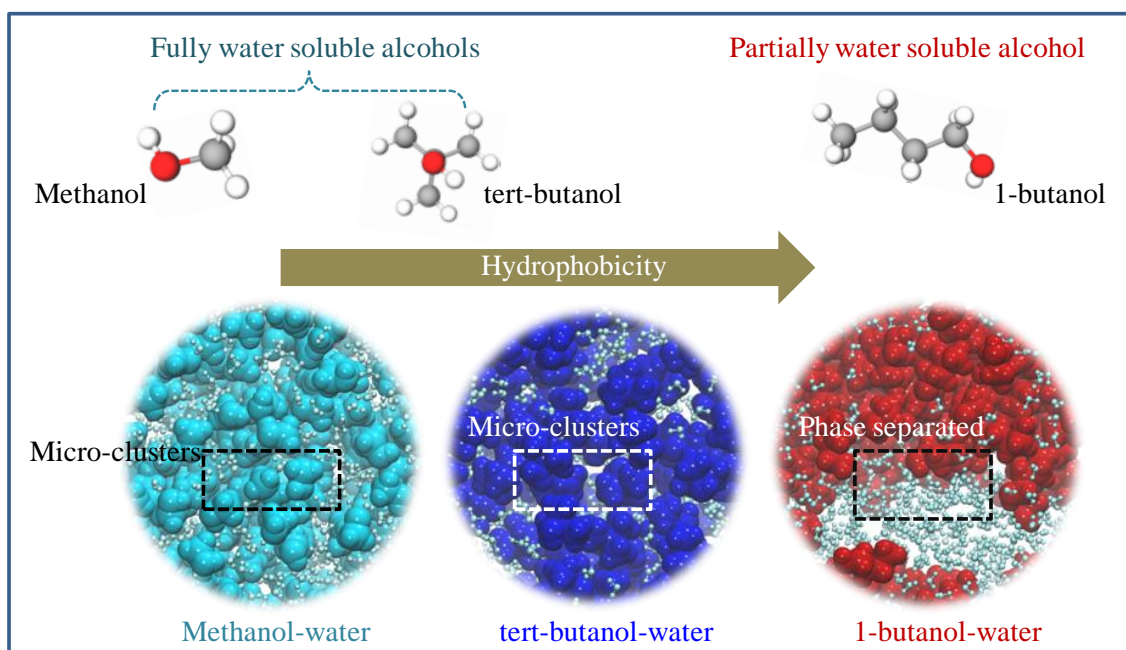
Scheme 1. The chemical structures of the alcohol molecules (methanol, tert-butanol, and 1-butanol) used in this study. MD simulation snapshots of the heterogeneous and phase-separated alcohol-water solutions.

2. Materials and Methods

Methanol (MOH), tert-butanol (TBA), and 1-butanol (1BA) were sourced from Sigma Aldrich and utilized in their as-received state without additional purification. The binary mixtures of alcohol and water were prepared using Milli-Q water, where the desired volume

fractions of alcohol(s) were achieved at $\Phi_{alc} = 10\%, 30\%, 50\%,$ and 70%. All measurements were conducted at room temperature.

2.1. THz-FTIR measurements.

THz-FTIR measurements were conducted within the far infrared (FIR) spectral range of 50-600 cm^{-1} using a Vertex 70V Fourier transform infrared spectrometer (Bruker, Germany) with an attached DTGS detector. The spectrometer was coupled with an attenuated total reflection (ATR) configuration featuring a sample compartment housing a diamond crystal (refractive index = 2.41). Measurements were executed in the ATR mode. Before each measurement, a rigorous sample compartment was cleaned, followed by evacuation using a vacuum pump. Each measurement cycle involved an average of 128 scans, utilizing a resolution of 4 cm^{-1}. The resulting spectral data acquired through the ATR unit were converted into absorbance and the absorption coefficient. Spectral contribution arises explicitly from the solvation shell has been calculated by subtracting the concentration-scaled absorption coefficients of bulk water and bulk alcohol from the aqueous alcohol solutions [19, 30, 33]:

$$\alpha_{hyd}(\nu) = \alpha_{solution}(\nu) - \Lambda_{alc}\alpha_{alc}(\nu) - \Lambda_w \alpha_w(\nu) \tag{1}$$

where, Λ_x is a concentration correction factor: $\mathbf{\Lambda_x} = \frac{c_{x,sol}}{c_{x,bulk}}$, $c_{x,sol}$ and $c_{x,bulk}$ are the alcohol (*alc*) and water (*w*) concentrations in the solutions and in its pure form at the same temperature, respectively. Subsequently, we fit the obtained $\boldsymbol{\alpha_{hyd}(\nu)}$ profiles summing up two damped harmonic oscillation models.

2.2. THz time-domain spectroscopy (TTDS).

THz time domain measurements were conducted using a commercially available THz spectrophotometer, TeraSmart, provided by Menlo Systems. A laser with a wavelength of 1560 nm and Erbium-doped fiber (ELMO by Menlo Systems) was utilized in the setup. All measurements were executed within a controlled nitrogen atmosphere with a humidity level below 10% at room temperature to circumvent the effects of water vapor absorption. This controlled environment was maintained using a liquid cell (Bruker, model A 145), equipped with z-cut quartz windows and a 100 μm thick Teflon spacer. The experimental process entailed altering the time delay between the probe and pump beams to record the amplitude and phase of the THz electric field as a function of time. Each experiment was replicated three times for accuracy. By employing Fourier analysis on the measured electric field amplitude ($E_{THz}(t)$), the frequency-dependent power and phase of the transmitted pulse were deduced. TTDS leverages coherent detection, enabling simultaneous measurement of both amplitude and phase in a single run. This method offers insights into the frequency-dependent optical properties of the system under study. The frequency-dependent absorption coefficient, $\boldsymbol{\alpha_{THz}(\nu)}$ and refractive index n(ν) can be obtained by the following equations:

$$\alpha_{THz}(\nu) = \frac{lnI_0(\nu) - lnI_S(\nu)}{d} \tag{2}$$

$$n(\nu) = \frac{\varphi_S(\nu) - \varphi_0(\nu)}{2\pi\nu d}c + 1 \tag{3}$$

where $I_0(\nu)$ and $I_S(\nu)$ are the intensity of the reference sample and alcohol-water solution respectively, $\varphi(\nu)$ specifies phase, and d is the path length. The frequency-dependent real (ε_{re}) and imaginary (ε_{im}) dielectric constants of the samples are obtained by following relevant

equations [34-36]. The water relaxation dynamics in the binary mixtures were extracted using the frequency-dependent complex dielectric constant, $\tilde{\varepsilon}(\nu) = \varepsilon_{re}(\nu) - i\varepsilon_{im}(\nu)$. The hydration dynamics has been extracted by fitting the complex dielectric profiles using multiple Debye relaxation models:

$$\tilde{\varepsilon}(\nu) = \varepsilon_\infty + \sum_{j=1}^{n} \frac{\varepsilon_j - \varepsilon_{j+1}}{1 + i2\pi\nu\tau_j} + \frac{\sigma}{i2\pi\nu\varepsilon_0} \qquad (4)$$

where τ_j is the relaxation time associated with the jth relaxation mode, ε_j are the dielectric constants, ε_1 represents the static dielectric constant, and ε_∞ is the dielectric constant extrapolated to very high frequency. σ is the solution conductivity (dc), and ε_0 is the permittivity in free space.

3. Results and Discussion

We measure the frequency-dependent absorption coefficient $\alpha(\nu)$ of the aqueous alcohol solutions in the FIR/THz frequency range (50-600 cm^{-1}) from the FTIR measurements. An illustrative $\alpha(\nu)$ profile for TBA-water binary mixtures at varying alcohol volume fractions (Φ_{alc}) is depicted in **Fig. 1a**. Pure water displays a distinct absorption feature within this frequency range owing to its coordinated movements and the intermolecular modes associated with HB stretching (around 130 cm^{-1}) and a constrained rotational librational mode (around 550 cm^{-1}). The $\alpha(\nu)$ profile decreases with the increase of alcohol content in the solution since the water molecules, which are the strong absorbers of THz frequency, are replaced by relatively less absorbing alcohol molecules. This phenomenon is commonly referred to as the "THz defect". This is a characteristic behavior observed with many hydrophobic solutes [19, 27, 34-36]. Notably, we have previously observed a distinctive bell-shaped non-ideality factor representing the deviation of the real absorption coefficient from the ideal case, with this effect becoming more pronounced in the higher frequency range and at higher alcohol[27]. This non-ideality might stem from creating defects within the HBond network of water, the aggregation of alcohol molecules, leading to alcohol-alcohol interactions, and possibly the development of clathrate-like structures. Alternatively, and more likely, it could result from a subtle interplay between these factors – defects in the HB network and alcohol-alcohol aggregation. The intricate nature of these interactions underscores the system's complexity under scrutiny.

To elucidate the role of pairwise intermolecular interactions contributing to this non-ideal behavior, we perform calculations of $\alpha_{hyd}(\nu)$ as per **Equ. 1**. In **Fig. 1b**, we present an illustrative $\alpha_{hyd}(\nu)$ plot for TBA. This parameter, by definition, is formulated by isolating the bulk contributions of both water and alcohol from the solution spectra. Consequently, $\alpha_{hyd}(\nu)$ ultimately becomes zero for any ideally mixed (non-interacting) combination. A distinct non-zero profile of $\alpha_{hyd}(\nu)$ robustly signifies the disruption of water's structure within the vicinity (i.e., the solvation shell) of the alcohol molecules. As a result, this parameter imparts explicit insights into the local solvation structural patterns.

As previously mentioned, specific isomers of butanol exhibit partial solubility in water, and upon surpassing certain threshold concentrations, they initiate a liquid-liquid phase separation phenomenon in binary aqueous mixtures [19, 30]. In instances where such phase-separated mixtures are encountered, we withdraw samples from the alcohol-rich phase(s) and proceed with measurements. When an amphiphilic molecule is dissolved in water, its hydrophilic portions interact with water through HBs. This interaction is manifest through the

onset of the librational mode in the spectra. Moreover, the water network arranges itself around and encases the hydrophobic section of the solute molecule, producing a distinctive spectral signature typically observed in the vicinity of ~190 cm^{-1} [37]. For a more quantified understanding, we subject the $\alpha_{hyd}(\nu)$ spectra to deconvolution using a sum of two damped harmonic oscillator (HO) models. This analytical approach enables us to extract detailed information about the structural characteristics and interactions within the system.

The fitted peak frequencies (ν_{HB} and ν_{Lib}) for these solutions, varying with Φ_{alc}, are visualized in **Fig. 1c** and **d**. Notably, ν_{HB} undergoes a gradual blue shift as Φ_{alc} increases, while ν_{Lib} exhibits a moderate redshift. This blue shift in ν_{HB} is attributed to the establishing of stronger HBs among water molecules that envelop the hydrophobic segments of alcohol molecules. This is also to note that an intriguing pattern exists in the observed increase of ν_{HB} with Φ_{alc}. While the change remains modest for water-soluble alcohols, it becomes substantially larger for partially soluble alcohols.
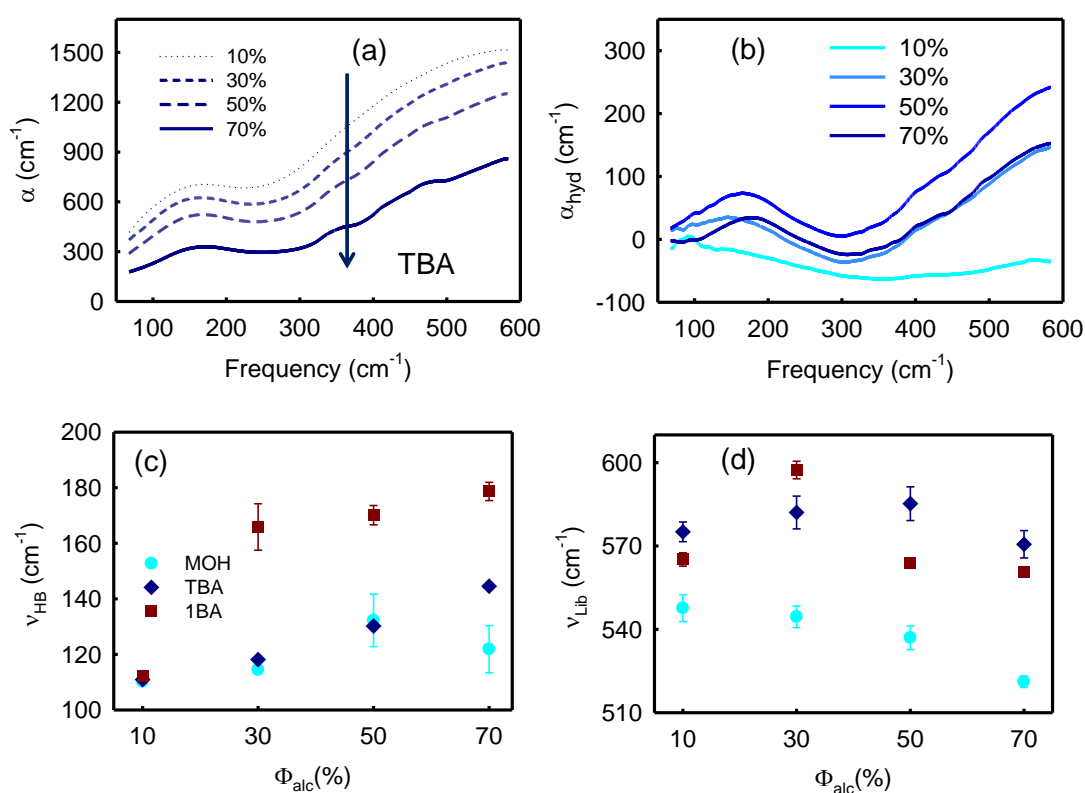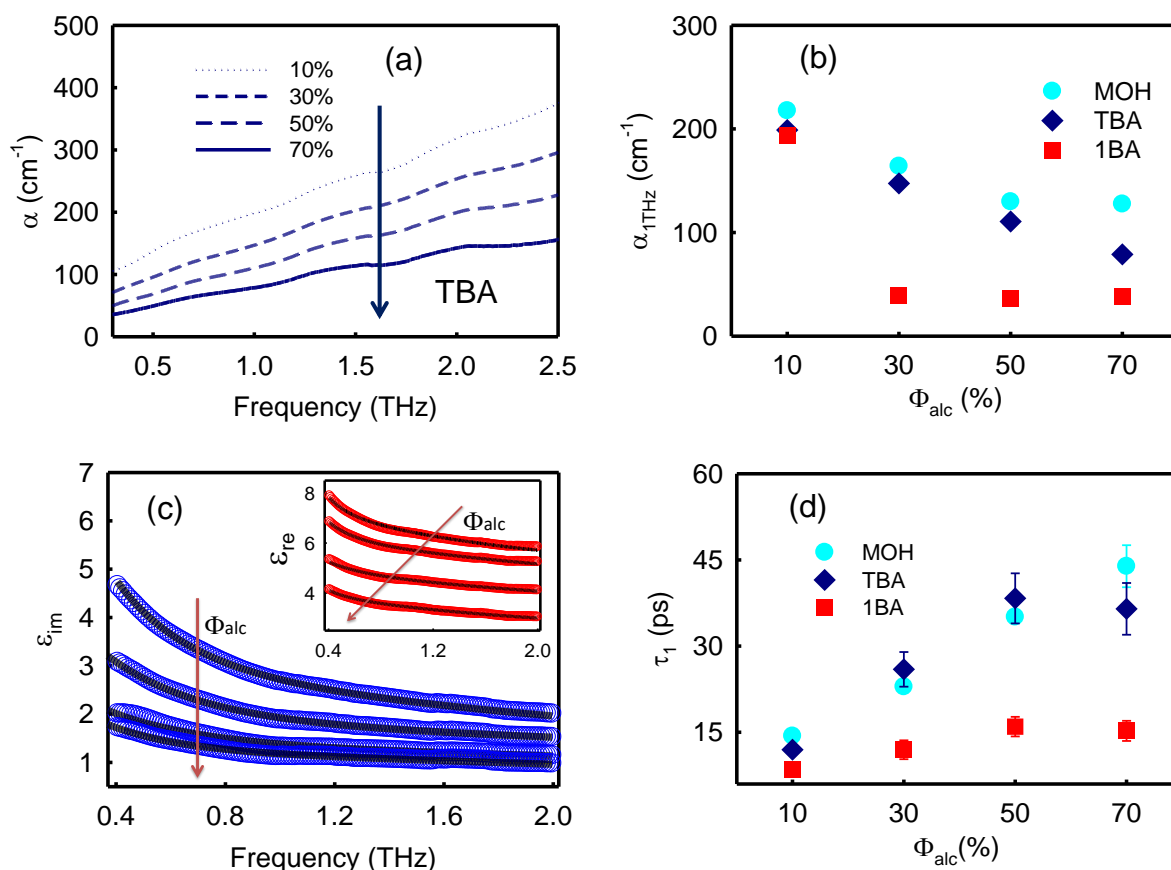

Figure 1. (a) THz absorption coefficients for TBA-water binary solutions. (b) Explicit contribution of solvation water for TBA-water solution. Dependency of the (c) central peak frequencies for the HBond stretching mode and (d) librational mode of the water in the aqueous-alcohol solutions with volume fractions.

The distinctions are notably clear: the MOH-water mixture displays the smallest blue shift in ν_{HB}. This outcome can be reasonably anticipated due to the lowest hydrophobicity and smallest size of methanol, enabling it to more seamlessly integrate into the water network and thus minimally perturb the HB structure. Following this trend, we also observe that the extent of the blue shift exhibits a linear escalation with the increased hydrophobicity of the alcohols. For the partially soluble alcohols, the conspicuous blue shift suggests *forming* a more robustly interconnected water network in the vicinity of the self-aggregated hydrophobic alcohols. An important note is that all the alcohol molecules employed in this study are known to display micro-heterogeneity and form self-associated molecular clusters of diverse dimensions in aqueous solutions *[16, 38-40]*. However, for water-soluble alcohols, the coexistence of

HBonded water clusters and self-assembled alcohol clusters is absent. This absence leads to the disruption of the HB network intrinsic to pure water, as it is disintegrated to accommodate the alcohol self-associated clusters. This particular phenomenon facilitates the mingling of the two liquids even at higher Φ_{alc}, despite the presence of alcohol molecule micro-clusters. In the context of aqueous binary mixtures involving 1BA, the observed phase separation can be attributed to the coexistence of alcohol self-associated clusters and HBonded water clusters[19, 32, 41]. The branched molecular structure of this isomer prompts a preference for forming elongated self-associations. This preference *minimizes* the contact area between these branched molecules and the surrounding water networks.

As previously highlighted, TTDS measurements offer the capability to elucidate diverse optical and dielectric properties inherent to water-alcohol mixtures, as summarized in Fig. 2. The parameter $\alpha(\nu_{THz})$ essentially signifies the combined dynamics of hydration exhibited by water molecules. In this context, a representative $\alpha(\nu)$ profile for TBA-water binary mixtures is depicted in Fig. 2a. The gradual decrease of $\alpha(\nu)$ with the augmentation of alcohol content is readily comprehensible. This phenomenon can be attributed to the replacement of water, with its higher THz absorption, by alcohol molecules exhibiting lower absorption characteristics [19, 27, 30]. For enhanced clarity, the α values at 1 THz are plotted against alcohol concentration in Fig. 2b. As anticipated, α decreases progressively in accordance with the increased size and hydrophobicity of the alcohol(s). Notably, at higher alcohol concentrations, the partially water-soluble alcohol (1BA) exhibits conspicuously lower absorption levels. This observation aligns with the characteristic behavior of the alcohol-enriched phase within liquid-liquid phase-separated water-alcohol binary mixtures. This trend is in line with similar observations made within the far-IR region [19].

Frequency-dependent real and imaginary dielectric constants decay profiles of water-TBA mixtures are presented in Fig. 2c. Pure water shows three distinct time constants of approximately 8 ps, around 200 fs, and roughly 80 fs. These values align well with findings from previous studies [19, 34-36]. The time constant of around 8 ps within the water is indicative of the spontaneous restructuring occurring within the HB-network [34, 36, 42]. The approximately 200 fs time likely arises from rapid jumps involving under-coordinated water or small angular rotations that precede larger angular jumps[43-44]. On the other hand, the ~80 fs time constant can be attributed to the 60 cm^{-1} vibrational band occurring due to the HB bending and related transverse acoustic phonons that propagate in a direction perpendicular to the HBs between adjacent water molecules [45]. Of note, previous reports have indicated that the reorientation of an individual alcohol monomer situated at the end of an alcohol-alcohol chain corresponds to a timescale of ~10 ps [46]. In alignment with these previous findings, we, too, have extracted a single relaxation mode with a picosecond timescale for concentrated alcohol solutions [20]. Our preceding study has already established that the behavior of (de)mixing within alcohol-water binary mixtures is intricately governed by the interplay between hydrophilic and hydrophobic hydration [19]. In light of this context, our study specifically focuses on the slow relaxation timescale (τ_1). This timescale effectively encapsulates collective information about alcohol solvation. As illustrated in Fig. 2d, τ_1 experiences a sharp increase with Φ_{alc}, and the magnitude of this increase is contingent upon the carbon chain length of the alcohol molecules. However, for partially water-soluble alcohol, particularly under phase-separated conditions - the HB relaxation is notably subtle, as depicted in Fig. 2d.

Figure 2. (a) THz absorption coefficient (α) of the water-TBA solutions. TBA content is indicated with a arrow. (b) α measured at 1 THz as a function of Φ_{alc} for different alcohols. (c) Real (ε_{re}) (in the inset) and imaginary (ε_{im}) dielectric constant of aqueous alcohol mixtures at different volume fractions in the THz frequency region. Solid lines indicate the multiple Debye relaxation fittings. (d) Dependency of the Debye relaxation time scale (τ_1) with Φ_{alc}.

The observed relaxation dynamics can be understood within the framework of a "wait-and-switch" model [47]. According to this model, the slow relaxation time constant (τ_1) indicates the time required for the reorganization of the HB network. As alcohol concentration in the mixtures increases, the available sites for new HBonding diminish, resulting in a heightened potential energy barrier between the two dipolar orientations of water. Consequently, the activation energy needed for molecular rotation becomes less likely to be surmounted, leading to a slower relaxation time constant. Interestingly, partially soluble butanol isomer 1BA displays an intriguingly faster relaxation despite having longer hydrophobic moieties compared to water-soluble alcohols. This phenomenon can be attributed to the relative dominance of molecular networks within the alcohol-water binary mixtures. Such rapid relaxation times, almost reminiscent of bulk-like behavior, signify the presence of a weak cross-molecular network within the alcohol-enriched portion of liquid-liquid phase-separated solutions.

On the contrary, within water-soluble alcohol solutions, the extended HBonded alcohol/water network contributes to an overall slowing-down of relaxation dynamics. Furthermore, the increase in the relaxation time constant has also been found to be accompanied by an enhanced radial distribution function (RDF). *This characteristic* has been interpreted as a collective inter-species cross-correlation. This interpretation aligns with the observations for water-soluble alcohols, and it has been confirmed through the HB stretching mode in FIR spectra, as illustrated in Fig. 1. Within partially soluble alcohols, the robustly

wrapped-around water networks originate from hydrophobic hydration and are notably stronger compared to those found in water-soluble alcohols.

4. Conclusions

With a comprehensive amalgamation of THz-TDS and THz-FTIR spectroscopy, this present contribution uncovers unique solvation behaviors within solute-solvated and solute-separated (micro) phases. The orientational relaxation dynamics of water emerge as a phenomenon intricately regulated by the relative prevalence of molecular networks within alcohol-water binary mixtures. Notably, butanol isomers prefer hydrophobic interactions, leading them to form self-aggregated molecular structures with relatively modest cross-molecular interactions. These molecular associations are, in turn, surrounded by water molecules engaged in water-water HB connectivity. Remarkably, relaxation dynamics akin to those within the bulk are observed within water networks proximate to aggregated alcohols. Conversely, alcohols with smaller hydrophobic chains engender solute-solvent extended HBonded networks, producing slower orientational dynamics. This distinctive relaxation dynamics in partially soluble alcohols is attributed to feeble cross-molecular HB interactions, yielding solute-enriched phases. This contrast in solvation dynamics implies the existence of self-aggregated and micro-heterogeneous phases within alcohol-water mixtures. Our study confirms that the hydrogen-bonded structure and its dynamics in alcohol-water binary mixtures governed by the fusion of hydrophobic chain length size and the solute aggregation-driven phase behavior within these mixtures.

Funding

Acknowledgments

Conflicts of Interest

The authors declare no conflict of interest.

References

1. Laage, D.; Elsaesser, T.; Hynes, J. T., Water dynamics in the hydration shells of biomolecules. *Chem. Rev.* **2017,** *117* (16), 10694-10725, doi.org/10.1021/acs.chemrev.6b00765.
2. Ball, P., Water as an active constituent in cell biology. *Chem. Rev.* **2008,** *108* (1), 74-108, doi.org/10.1021/cr068037a.
3. Böhmer, R.; Gainaru, C.; Richert, R., Structure and dynamics of monohydroxy alcohols—Milestones towards their microscopic understanding, 100 years after Debye. *Phys. Rep.* **2014,** *545* (4), 125-195, doi.org/10.1016/j.physrep.2014.07.005.
4. Dzida, M.; Kaatze, U., Compressibility and dielectric relaxation of mixtures of water with monohydroxy alcohols. *J. Phys. Chem. B* **2015,** *119* (38), 12480-12489, doi.org/10.1021/acs.jpcb.5b07093.
5. Zhang, X.; Zhu, Y.; Granick, S., Hydrophobicity at a Janus interface. *Science* **2002,** *295* (5555), 663-666, DOI: 10.1126/science.106614.
6. Böhm, F.; Schwaab, G.; Havenith, M., Mapping Hydration Water around Alcohol Chains by THz Calorimetry. *Angewandte Chemie International Edition* **2017,** *56* (33), 9981-9985, https://doi.org/10.1002/anie.201612162.

7. Fukasawa, T.; Amo, Y.; Tominaga, Y., Low-frequency Raman scattering study of tert-butyl alcohol-water and tetrahydrofuran-water binary mixtures. *Journal of Chemical Physics* **2003,** *118* (14), 6387-6393, 10.1063/1.1559672.

8. Dougan, L.; Hargreaves, R.; Bates, S. P.; Finney, J. L.; Ŕat, V.; Soper, A. K.; Crain, J., Segregation in aqueous methanol enhanced by cooling and compression. *Journal of Chemical Physics* **2005,** *122* (17), 10.1063/1.1888405.

9. Mizuno, K.; Kimura, Y.; Morichika, H.; Nishimura, Y.; Shimada, S.; Maeda, S.; Imafuji, S.; Ochi, T., Hydrophobic hydration of tert-butyl alcohol probed by NMR and IR. *Journal of Molecular Liquids* **2000,** *85* (1), 139-152, https://doi.org/10.1016/S0167-7322(99)00170-1.

10. Andersen, J.; Heimdal, J.; Wugt Larsen, R., The influence of large-amplitude librational motion on the hydrogen bond energy for alcohol–water complexes. *Physical Chemistry Chemical Physics* **2015,** *17* (37), 23761-23769, 10.1039/C5CP04321B.

11. Majumdar, B. B.; Pyne, P.; Mitra, R. K.; Das Mahanta, D., Impact of hydrophobicity on local solvation structures and its connection with the global solubilization thermodynamics of amphiphilic molecules. *Physical Chemistry Chemical Physics* **2023,** *25* (40), 27161-27169, 10.1039/D3CP02741D.

12. Wakisaka, A.; Komatsu, S.; Usui, Y., Solute-solvent and solvent-solvent interactions evaluated through clusters isolated from solutions: Preferential solvation in water-alcohol mixtures. *Journal of Molecular Liquids* **2001,** *90* (1-3), 175-184, doi.org/10.1016/S0167-7322(01)00120-9.

13. Sarkar, S.; Saha, D.; Banerjee, S.; Mukherjee, A.; Mandal, P., Broadband terahertz dielectric spectroscopy of alcohols. *Chemical Physics Letters* **2017,** *678*, 65-71, https://doi.org/10.1016/j.cplett.2017.04.026.

14. Pyne, P.; Das Mahanta, D.; Gohil, H.; Prabhu, S.; Mitra, R. K., Correlating solvation with conformational pathways of proteins in alcohol–water mixtures: a THz spectroscopic insight. *Physical Chemistry Chemical Physics* **2021,** *23* (32), 17536-17544, doi.org/10.1039/D1CP01841H.

15. Kölbel, J.; Schirmacher, W.; Shalaev, E.; Zeitler, J. A., Terahertz dynamics in the glycerol-water system. *Physical Review B* **2023,** *107* (10), 104203, 10.1103/PhysRevB.107.104203.

16. Banerjee, S.; Furtado, J.; Bagchi, B., Fluctuating micro-heterogeneity in water–tert-butyl alcohol mixtures and lambda-type divergence of the mean cluster size with phase transition-like multiple anomalies. *The Journal of Chemical Physics* **2014,** *140* (19), 194502, http://dx.doi.org/10.1063/1.4874637.

17. Pérez, E. G.; González-Salgado, D.; Lomba, E., Molecular dynamics simulations of aqueous solutions of short chain alcohols. Excess properties and the temperature of maximum density. *Fluid Phase Equilibria* **2021,** *528*, 112840, https://doi.org/10.1016/j.fluid.2020.112840.

18. Urbina, A. S.; Slipchenko, L. V.; Ben-Amotz, D., Quantifying the Nearly Random Microheterogeneity of Aqueous tert-Butyl Alcohol Solutions Using Vibrational Spectroscopy. *The Journal of Physical Chemistry Letters* **2023,** *14* (50), 11376-11383, 10.1021/acs.jpclett.3c02603.

19. Chakraborty, S.; Pyne, P.; Kumar Mitra, R.; Das Mahanta, D., A subtle interplay between hydrophilic and hydrophobic hydration governs butanol (de)mixing in water. *Chemical Physics Letters* **2022,** *807*, 140080, https://doi.org/10.1016/j.cplett.2022.140080.

20. Li, R.; D'Agostino, C.; McGregor, J.; Mantle, M. D.; Zeitler, J. A.; Gladden, L. F., Mesoscopic Structuring and Dynamics of Alcohol/Water Solutions Probed by Terahertz Time-Domain Spectroscopy and Pulsed Field Gradient Nuclear Magnetic Resonance. *The Journal of Physical Chemistry B* **2014,** *118* (34), 10156-10166, 10.1021/jp502799x.

21. Pyne, P.; Pyne, S.; Mitra, R. K., Sugar Molecules Inhibit Insulin Aggregation: A Decisive Role Being Played by the Protein Solvation Energetics. *The Journal of Physical Chemistry B* **2023,** *127* (41), 8825-8832, 10.1021/acs.jpcb.3c04765.

22. Saha, R.; Mitra, R. K., Thermo-Resistive Phase Behavior of Trivalent Ion-Induced Microscopic Protein-Rich Phases: Correlating with Ion-Specific Protein Hydration. *Langmuir* **2023,** *39* (13), 4601-4610, 10.1021/acs.langmuir.2c03302.

23. Bag, S.; Pezzotti, S.; Das Mahanta, D.; Schulke, S.; Schwaab, G.; Havenith, M., From Local Hydration Motifs in Aqueous Acetic Acid Solutions to Macroscopic Mixing Thermodynamics: A Quantitative Connection from THz-Calorimetry. *The Journal of Physical Chemistry B* **2023,** *127* (42), 9204-9210, 10.1021/acs.jpcb.3c06328.

24. Mazaheri, Z.; Papari, G. P.; Andreone, A., Probing the Molecular Dynamics of Aqueous Binary Solutions with THz Time-Domain Ellipsometry. *Sensors* **2023,** *23* (4), 2292, doi.org/10.3390/s23042292.

25. Das Mahanta, D.; Islam, S. I.; Choudhury, S.; Das, D. K.; Mitra, R. K.; Barman, A., Contrasting hydration dynamics in DME and DMSO aqueous solutions: A combined optical pump-probe and GHz-THz dielectric relaxation investigation. *Journal of Molecular Liquids* **2019,** *290*, 111194, https://doi.org/10.1016/j.molliq.2019.111194.

26. Samanta, N.; Das Mahanta, D.; Mitra, R. K., Collective hydration dynamics of guanidinium chloride solutions and its possible role in protein denaturation: a terahertz spectroscopic study. *Phys. Chem. Chem. Phys.* **2014,** *16* (42), 23308-23315, 10.1039/C4CP03273J.

27. Das, D. K.; Das Mahanta, D.; Mitra, R. K., Nonmonotonic Hydration Behavior of Bovine Serum Albumin in Alcohol/Water Binary Mixtures: A Terahertz Spectroscopic Investigation. *ChemPhysChem* **2017,** *18* (7), 749-754, 10.1002/cphc.201601217.

28. Kindt, J. T.; Schmuttenmaer, C. A., Far-infrared dielectric properties of polar liquids probed by femtosecond terahertz pulse spectroscopy. *J. Phys. Chem.* **1996,** *100*, 10373-10379, doi.org/10.1021/jp960141g.

29. Yomogida, Y.; Sato, Y.; Nozaki, R.; Mishina, T.; Nakahara, J. i., Comparative dielectric study of monohydric alcohols with terahertz time-domain spectroscopy. *Journal of Molecular Structure* **2010,** *981* (1), 173-178, https://doi.org/10.1016/j.molstruc.2010.08.002.

30. Chakraborty, S.; Pyne, P.; Mitra, R. K.; Das Mahanta, D., Hydrogen bond structure and associated dynamics in micro-heterogeneous and in phase separated alcohol-water binary mixtures: A THz spectroscopic investigation. *Journal of Molecular Liquids* **2023,** *382*, 121998, https://doi.org/10.1016/j.molliq.2023.121998.

31. Saha, R.; Mitra, R. K., Trivalent cation-induced phase separation in proteins: ion specific contribution in hydration also counts. *Physical Chemistry Chemical Physics* **2022,** *24* (38), 23661-23668, 10.1039/D2CP01061E.

32. Choi, S.; Parameswaran, S.; Choi, J.-H., Effects of molecular shape on alcohol aggregation and water hydrogen bond network behavior in butanol isomer solutions. *Physical Chemistry Chemical Physics* **2021,** *23* (23), 12976-12987, doi.org/10.1039/D1CP00634G.

33. Das Mahanta, D.; Brown, D. R.; Pezzotti, S.; Han, S.; Schwaab, G.; Shell, M. S.; Havenith, M., Local solvation structures govern the mixing thermodynamics of glycerol–water solutions. *Chemical Science* **2023,** *14* (26), 7381-7392, 10.1039/D3SC00517H.

34. Das Mahanta, D.; Samanta, N.; Mitra, R. K., Decisive Role of Hydrophobicity on the Effect of Alkylammonium Chlorides on Protein Stability: A Terahertz Spectroscopic Finding. *J. Phys. Chem. B* **2017,** *121* (33), 7777-7785, 10.1021/acs.jpcb.7b04088.

35. Das Mahanta, D.; Samanta, N.; Mitra, R. K., The effect of monovalent cations on the collective dynamics of water and on a model protein. *J. Mol. Liq.* **2016,** *215*, 197-203, http://dx.doi.org/10.1016/j.molliq.2015.12.031.

36. Das Mahanta, D.; Patra, A.; Samanta, N.; Luong, T. Q.; Mukherjee, B.; Mitra, R. K., Non-monotonic dynamics of water in its binary mixture with 1,2-dimethoxy ethane: A combined THz spectroscopic and MD simulation study. *J. Chem. Phys.* **2016,** *145* (16), 164501, http://dx.doi.org/10.1063/1.4964857.

37. Conti Nibali, V.; Pezzotti, S.; Sebastiani, F.; Galimberti, D. R.; Schwaab, G.; Heyden, M.; Gaigeot, M. P.; Havenith, M., Wrapping Up Hydrophobic Hydration: Locality Matters. *The Journal of Physical Chemistry Letters* **2020,** *11* (12), 4809-4816, 10.1021/acs.jpclett.0c00846.

38. Wakisaka, A.; Matsuura, K., Microheterogeneity of ethanol–water binary mixtures observed at the cluster level. *Journal of Molecular Liquids* **2006,** *129* (1), 25-32, https://doi.org/10.1016/j.molliq.2006.08.010.

39. Tomza, P.; Czarnecki, M. A., Microheterogeneity in binary mixtures of propyl alcohols with water: NIR spectroscopic, two-dimensional correlation and multivariate curve resolution study. *Journal of Molecular Liquids* **2015,** *209*, 115-120, https://doi.org/10.1016/j.molliq.2015.05.033.

40. Penkov, N. V., Application of Terahertz Time-Domain Spectroscopy to Study the Microheterogeneities of Solutions: A Case Study of Aqueous Sugar Solutions. *Photonics* **2023,** *10* (8), 887, doi.org/10.3390/photonics10080887.

41. Das Mahanta, D.; Rana, D.; Patra, A.; Mukherjee, B.; Mitra, R. K., Heterogeneous structure and solvation dynamics of DME/water binary mixtures: A combined spectroscopic and simulation investigation. *Chem. Phys. Lett.* **2018,** *700*, 50-56, https://doi.org/10.1016/j.cplett.2018.04.003.

42. van der Post, S. T.; Tielrooij, K.-J.; Hunger, J.; Backus, E. H. G.; Bakker, H. J., Femtosecond study of the effects of ions and hydrophobes on the dynamics of water. *Faraday Discussions* **2013,** *160* (0), 171-189, 10.1039/C2FD20097J.

43. Laage, D.; Hynes, J. T., A Molecular Jump Mechanism of Water Reorientation *Science* **2006,** *311*, 832-835, DOI: 10.1126/science.1122154.

44. Das Mahanta, D.; Mitra, R. K., Connection of large amplitude angular jump motions with temporal heterogeneity in aqueous solutions. *Physical Chemistry Chemical Physics* **2020,** *22* (17), 9339-9348, 10.1039/D0CP00491J.

45. Vij, J. K.; Simpson, D. R. J.; Panarina, O. E., Far infrared spectroscopy of water at different temperatures: GHz to THz dielectric spectroscopy of water *J. Mol. Liq.* **2004,** *112*, 125-135, doi.org/10.1016/j.molliq.2003.12.014.

46. Venables, D. S.; Schmuttenmaer, C. A., Far-infrared spectra and associated dynamics in acetonitrile-water mixtures measured with femtosecond THz pulse spectroscopy. *J. Chem. Phys.* **1998,** *108*, 4935-4943, doi.org/10.1063/1.475902.

47. Petong, P.; Pottel, R.; Kaatze, U., Dielectric Relaxation of H-Bonded Liquids. Mixtures of Ethanol and n-Hexanol at Different Compositions and Temperatures. *The Journal of Physical Chemistry A* **1999,** *103* (31), 6114-6121, 10.1021/jp991046l.