I. INTRODUCTION

Water is the most abundant and the principle constituent of any living organism. In biological systems, water is confined between biomolecular assemblies where water dynamics is relevant in membrane functioning and cytoskeletal organizations. Recently, protein displacements in live cell membranes are found to have robust exponential tails due to the underlying dynamical heterogeneities. The nano-scale heterogeneity in membrane dynamics is believed to play the dominant role in various cellular processes such as signal transduction, matter transport and enzymatic activities in cells. Dynamical heterogeneities in fluid membranes are observed on the scale of 80-150 nm with super-resolution stimulated emission depletion microscopic and fluorescence correlation spectroscopic techniques, even in absence of cholesterols.

Although, dynamical heterogeneity in membranes is not well-studied yet, the obvious question comes if dynamical heterogeneity in the membrane is coupled with the dynamics of its hydration layer. In the last decade, with a major advancement in computer simulation techniques, water near soft interfaces are found to have very slow relaxation times. However, the origin and mechanism of slow relaxation time have not been analyzed in a systematic manner with microscopic details till date. If universal slow hydration dynamics is due to the breaking of tetrahedral hydrogen bond network near interfaces, is their dynamics dependent on the chemical nature of the confinement present in the interfaces? Confined diffusion of nano-particles are found to follow non-gaussian statistics even at the long time Brownian stage. Dynamical heterogeneity time and length scales are analyzed for the ionic liquids and water binary mixtures where jump motions are evident from the non-gaussian distributions. Activated hopping is known to facilitate the random diffusion in supercooled liquids. Water confined near protein surface are shown to exhibit a dynamic crossover with a breakdown of Stokes Einstein relation at supercooled temperatures. Water confined near silica hydrophilic pores at room temperature reveal two different dynamical regimes similar to supercooled bulk water. The open question remains, do confined water at room temperature dynamically behave very similar to supercooled bulk water? If yes, what are the sources of such distinct dynamics?

In this article we present evidences of dynamical heterogeneities in chemically confined water near DMPC lipid headgroups at temperature (308 K) well above supercooling. The influence of chemical nature on the hydrogen bond dynamics of interface water is systematically analyzed to find out the origin of their slow relaxation rates. Our calculations provide the microscopic mechanism responsible for such behavior at room temperature with their implications and potential applications.

II. SIMULATION DETAILS

An all atom molecular dynamics simulation is carried out for 128 DMPC molecules in presence of 5743 TIP4P/2005 water molecules using previously equilibrated DMPC system at 308 K. Force field parameters for DMPC are obtained using Berger united atom force field. An NPT run is carried out for 100 ns with a 2 fs time step. The system is equilibrated at 308 K using velocity rescaling method with a coupling constant of 0.5 ps. The pressure is maintained at 1 bar using semi-isotropic pressure coupling by Berendsen...
pressure coupling with a coupling constant of 0.1 ps. Coulombic and van der Waal interactions were cutoff at 1 nm. Long range interactions are corrected using particle mesh Ewald method with a 4 nm grid size. Periodic boundary conditions are applied in all three directions. An NVT simulation is performed for 1.9 ns with a 0.4 fs time step where the last 1 ns run is analyzed for water dynamics. Parameters for temperature coupling, cutoff distances and long range interactions are kept same as in the previous run. Trajectories are collected at every 10 fs. The simulation box-length for the hydrated DMPC lipid is 6.24 nm along x and y directions and 7.95 nm along z direction. To compare the dynamics of interfacial water with bulk water (BW), a NVT run is carried out for a box of 851 TIP4P/2005 water molecules using the same parameters as in DMPC-water system. For 2 ns with a 2 fs time step in an NPT ensemble with the same set of parameters as in the hydrated DMPC. Next a NVT run is carried out for 200 ps with a 0.4 fs time step. The 100 ps NVT run is further extended till 200 ps where we have used last 100 ps data for analysis. The box length for BW is 3.69 nm along x and y and 1.84 nm along z directions. Trajectories are collected at every 10 fs. All simulations are carried out using Gromacs 4.6,52,53–56.

Figure 1 (a) and (b) show the bilayer and a single DMPC molecule present in the bilayer respectively. To decouple the contribution of bulk water (BW) to the dynamical properties of hydration layers, interface water are classified based on geometric definitions. If a water molecule continuously resides in a layer which is ±3Å away from the location of the head group density of DMPC, the molecule is identified as interface water (IW).52,59. The inset in figure 2 shows the number of IW with respect to their lifetimes. IW has a bi-exponential lifetime dependence which is indicative of the presence of two characteristic relaxation time-scales. IW which stay continuously in the hydration layer for 100 ps are found to provide reasonable statistics to calculate dynamical quantities and used for further analyses. If one IW of 100 ps lifetime is hydrogen bonded to another IW with same lifetime, it is referred to as IW-IW. Additionally, if one pair of IW-IW is concerted hydrogen bonded to the carbonyl (CO), phosphate (PO) or glycerol (Glyc) moiety of lipid heads (shown in different colors in figure 1(b)), the IW-IW is referred to as IW-CO, IW-PO or IW-Glyc respectively.52. The approach clearly deconstructs the contributions of different chemical environments on the dynamical behaviors of interface water which is lacking if water molecules are characterized depending on their vertical positions along the bilayer normal. To obtain the dynamics of the slowest IW with reasonable statistics, another class of IW is chosen which stay continuously in the hydration layer for 400 ps and is referred to as IW$_{CR400}$

### III. RESULTS AND DISCUSSIONS

The radial distribution functions ($g(r)$) between two oxygen atoms and oxygen and hydrogen atoms of different classes of IW are shown in figure 2(a) and (b) respectively. The highest amplitude of IW$_{CR400}$ with respect to remaining classes of water indicates that the IW$_{CR400}$ have the lowest potential energies due to the breakdown of tetrahedral networks while forming hydrogen bonds to the lipid head moieties. To analyze the dynamical nature of bound waters, translational mean square displacement (MSD, $\frac{1}{N} \sum_{i=1}^{N} <(r_i(t) - r_i(0))^2>$) is calculated for all classes of water. Figure 3(a) shows that the BW follows diffusive behavior at longer time where all classes of IW remain sub-diffusive due to the trapping in a cage formed by the neighboring hydrogen bond networks. IW$_{CR400}$ exhibit the slowest MSD due to the longest confinement lifetime. To understand the origin of subdiffusive nature of the IW, non-Gaussian parameter (NGP, $\alpha_2$) are calculated using the equation, $\alpha_2(t) = \frac{3\Delta r(t)^2}{5\Delta r(t)^2} - 1$ (figure 3). The NGP for the BW reaches a maximum within a small time, 0.9 ps and then decays asymptotically to zero confirming the gaussian diffusion. The values of NGP for different classes of IW, start increasing to much higher amplitudes than that for the BW and reach to maxima in between $\beta$ and $\alpha$-relaxation time scales. The slow decays after the peak of NGP are due to the release of the IW from the respective cages via diffusion. Similar cross-over from cage to translational diffusive regime at the peak of NGP are found for Brownian particles in a periodic effective field53. Since different classes of IW which continuously reside for 100 ps in the hydration layer, leave the layer after their confinement lifetimes, the maxima of their respective NGP are nearly at 100 ps (see Table I) IW-Glyc being burried deepest in the hydrophobic core of lipid (figure 1(b)) show the highest $\beta$-relaxation among IW which are continuously residing for 100 ps. Interestingly, IW$_{CR400}$ exhibit very strong non-gaussian behavior for longer period of time compared to other classes of water molecules and decay after 340 ps which is again close to their confinement lifetimes. A very slow $\alpha$-relaxation of IW$_{CR400}$ is indicative of their slow diffusion towards far-interface region. The broad $\beta$-relaxations of all classes of IW signify a strong structural arrest by their surrounding molecules similar to supercooled liquids exhibiting dynamical heterogeneity2,34. The transition from the $\beta$-relaxation to the $\alpha$-relaxation occurs at similar spatio-temporal scale where the respective MSD leave the sub-diffusive regime.

| Region | $\tau$ (ps) |
|--------|-------------|
| IW     | 78.70       |
| IW-IW  | 70.10       |
| IW-CO  | 80.10       |
| IW-Glyc| 82.00       |
| IW-PO  | 78.40       |
| IW$_{CR400}$ | 344.80 |
| BW     | 0.90        |

TABLE I. Relaxation time-scales of different IW and BW as obtained from NGP. For BW, $\alpha_2$ decays to 0 following Fickian dynamics where remaining IW show strong non-gaussian behavior.
FIG. 1. a) Snapshot of a DMPC bilayer in presence of interface water, b) snapshot of a single DMPC molecule showing different moieties in different colors, blue: carbonyl carbon, red: oxygen, green: phosphorus of phosphate, violet: glycerol carbon.

FIG. 2. RDF for different classes of IW and BW between a) oxygen-oxygen and b) oxygen-hydrogen atoms. Inset: Lifetime of interfacial waters continuously residing (CR) within ±3Å away from the peak of the density profile of nitrogen atoms of lipid heads.

FIG. 3. a) Translational mean square displacement, b) NGP for all classes of IW and BW show a crossover from β-relaxation to α-relaxation at the same time-scale when respective sub-diffusive regimes are left for diffusion.

FIG. 4. Translational self part of van Hove correlation function for all classes of IW and BW. All IW pertain large length correlation than the BW. Inset: van Hove correlation function along x direction for all classes of IW and BW. IW show a strong deviation from gaussianity.

For gaining deeper insights in the dynamical evolution of IW associated with lipid moieties, self part of radial van Hove correlation function is calculated via,

\[ G_s(r, t) = \frac{1}{N} \sum_{i=1}^{N} \delta(r + r_i(0) - r_i(t)) \]

The inset in the figure represents one dimensional van Hove correlation function where the BW follow gaussian dynamics and all classes of IW show larger deviations from gaussianity via exponential tails. Similar exponential tails have been manifested as an established behavior of dynamical heterogeneity in supercooled glass forming liquids.

Since self intermediate scattering function (SISF, \( F_s(q, t) \)) is another universal feature of dynamical...
heterogeneity, two dimensional \( F_s(q,t) \) is calculated by,

\[
F_s(q,t) = \frac{1}{\lambda} \left( \sum_{i=1}^{N} \cos(q, r_i(t) - r_i(0)) \right).
\]

\( q \) is obtained from the location of the first peak of the \( q(r) \) via \( q = \frac{2\pi n}{\lambda} \) where \( \lambda \) is the wave length. To obtain the wave-vector dependence on the \( \alpha \)-relaxation times, SIF is calculated at different values of \( \lambda \). Figure 5 (a) and (b) show the behavior of SIF at different \( \lambda \) for the IW\(_{CR400}\) and the BW. Although the \( \beta \) and \( \alpha \)-relaxation times of the IW and the BW are not prominently disparate by characteristics boson peaks as observed for the supercooled liquids. SIF very clearly scales up in different time regimes. The wave length dependence of \( \alpha \)-relaxation time-scales \( (\tau_\alpha) \) is generally characterized by the exponential decay of \( F_s(q,t) = \exp(-Dq^2 t) \) at all wave lengths at room temperature. If diffusion coefficients \( (D) \) and relaxation times of IW follow a distribution at room temperature due to the dynamical heterogeneity, the quadratic dependence of \( \lambda \) to the long relaxation time will not be followed anymore. To check that, the long \( t \) are extracted from the data in figure 5 (a) and (b) for the IW\(_{CR400}\) and the BW respectively. The inset in figure 5 (c) shows much slower \( \alpha \)-relaxation time-scales for the IW\(_{CR400}\) in comparison to the BW (the data for the BW is multiplied by a factor of 5 for a better comparison). Interestingly, the \( \tau_\alpha \) of the BW follow quadratic dependence to the \( \lambda \) which changes to a linear dependence for the IW\(_{CR400}\). This is probably because the movement of confined water are heterogeneous in nature at all length-scales due to the breakdown of Stokes-Einstein relation at room temperature. \( \lambda = 0.5 \) nm is chosen to understand the nature of characteristics relaxation time-scales of all classes of IW and BW (figure 5 (c)). All IW which continuously reside at the hydration layer for 100 ps show much slower relaxation compare to the BW. Our BW data are fitted best to two relaxation time-scales using,

\[
F_s(q,t) = (1 - f_Q) \exp[-(\frac{q}{\lambda})^2] + f_Q \exp[-(\frac{q}{\lambda})^{\beta_\alpha}].
\]

\( f_Q \) is known as the Debeye-Waller factor, \( \tau_s \) is the time-scale for ballistic motion and \( \tau_\alpha \) is the relaxation time-scale of the cage. The stretched exponential in the equation is known as Kohlrausch-William-Watt \( (KWW) \) function \( \beta_\alpha \) \( \beta_\alpha \) can be correlated with the breakdown of Stokes-Einstein relation for supercooled liquids. However, all classes of IW are not fitted to the previous equation due to the appearance of a long time tail very similar to that for the BW near proteins. The SISF of IW can be accounted with three relaxation time-scales where one more stretched parameter is added to the KWW function as \( F_s(q,t) = (1 - f_Q - f_Q^1) \exp[-(\frac{q}{\lambda})^2] + f_Q \exp[-(\frac{q}{\lambda})^{\beta_\alpha}] + f_Q^1 \exp[-(\frac{q}{\lambda})^\beta_\alpha] \). \( \tau_1 \) and \( \beta_1 \) are longer relaxation time and stretching parameter respectively. Although such stretched long relaxation is not so common in glass-former liquids, \( \tau_1 \) for the IW near protein is known to follow Arrhenius dependence on temperatures with a cross-over due to the protein structure fluctuations.

Table 2 shows the relaxation time-scales and stretching parameters for all IW and the BW near the DMPC. \( \tau_\alpha \) and \( \tau_\alpha \) for all classes of IW which continuously reside in the hydration layer for 100 ps, show similar time-scales as that of the BW and consistent with the time-scales obtained for hydration water near protein or sugar. The IW-CO/IW-Glyc exhibit slower \( \tau_\alpha \) and \( \tau_\alpha \) since they are buried deepest in the hydrophobic region of lipids. Interestingly, the \( \tau_\alpha \) and \( \tau_\alpha \) for IW\(_{CR400}\) are 10 orders of magnitude slower than the remaining classes of water. However, all classes of water follow a long time stretched exponential tail (described by \( \tau_1 \) which is clearly absent in the BW. Notably, the \( \tau_1 \) for IW\(_{CR400}\) is 4-6 times larger than that for the remaining classes of IW. The emergence of \( \tau_1 \) is attributed to the very slow relaxation of the IW arrested in a cage of hydrogen bond networks formed near the lipid head groups. Since the time-scale of \( \tau_1 \) for the IW matches with the reorientation and translation time-scale of lipid heads (which is 10-100 ns), a coupling between lipid head dynamics to the IW hydrogen bond dynamics can be the source of such long relaxations.

![Figure 5](image_url)

**FIG. 5.** Self intermediate scattering function, \( F_s(q,t) \) for a) IW\(_{CR400}\) and b) BW at different \( \lambda \) ranging from \( \lambda = 0.15 \) nm to \( \lambda = 3.00 \) nm \( (q = \frac{2\pi n}{\lambda}) \). Inset: \( \alpha \)-relaxation dependence on \( \lambda \). Symbols: \( \tau_\alpha \) and \( \tau_1 \) for IW\(_{CR400}\) and BW respectively; lines : fitting.

| Region   | \( \tau_\alpha \) | \( f_Q \) | \( \tau_\alpha \) | \( \beta_\alpha \) | \( f_Q^1 \) | \( \tau_1 \) | \( \beta_1 \) |
|----------|------------------|--------|------------------|------------------|--------|------------------|------------------|
| IW       | 0.29 0.23        | 2.87   | 0.99 0.71        | 15.26 0.48       |
| IW-IW    | 0.21 0.36        | 2.69   | 0.92 0.55        | 20.93 0.59       |
| IW-CO    | 0.32 0.21        | 2.86   | 0.99 0.75        | 15.03 0.46       |
| IW-Glyc  | 0.31 0.20        | 2.74   | 0.92 0.77        | 15.12 0.45       |
| IW-PO    | 0.29 0.26        | 2.69   | 0.91 0.69        | 15.15 0.48       |
| IW\(_{CR400}\) | 3.72 0.63       | 26.29 0.33 | 0.23 83.59 0.89   |
| BW       | 0.24 0.88        | 2.53   | 0.93             |                   |
IV. CONCLUSIONS

In summary, the article provides evidences of spatiotemporal heterogeneities in interface water near lipid membranes at temperature well above the glass transition temperature using all-atom molecular dynamics simulations. Since IW_{CR400} has the longest confinement lifetime, more profound dynamical heterogeneities are observed for these molecules. Although dynamical heterogeneities persist for all classes of IW near the lipid membranes, the magnitude of the heterogeneities are largely dictated by the hydrogen bond partner of lipid head moieties. IW-PO, IW-CO, IW-Glyc are interface water molecules which are hydrogen bonded among each other and conversely hydrogen bonded to PO, CO, Glyc of lipid heads. So for these cases, the dynamics are essentially probed for the hydrogen bonds between two IW molecules, although the hydrogen bond partners are different for IW-CO to IW-PO to IW-Glyc. Thus, the chemical nature of the PO, CO or Glyc of lipid heads have less influence on the IW-IW hydrogen bond dynamics. On the other hand, IW_{CR400} include only those interface water which have formed hydrogen bonds directly to the lipid head moieties: PO, CO, Glyc and remain intact for the entire 400 ps confinement lifetime. Thus the lipid partners have more influences on the dynamics of the respective IW_{CR400}. Since these IW have formed hydrogen bonds to PO or CO or Glyc simultaneously, their interactions are more heterogeneous in nature. The heterogeneous interactions within IW_{CR400} may lower down the potential energy of the respective cages compared to the ones having the homogeneous interactions very similar to the supercooled Lennard-Jones liquids. The heterogeneous interactions of IW_{CR400} can generate a distribution of relaxation times which might be slower than the IW of remaining kind. Thus our calculations reveal for the first time that the slow relaxations of chemically confined water molecules near lipid membranes are originated from dynamical heterogeneities at a temperature well above supercooling.

Additionally, our analysis shows strong signatures of coupling of lipid dynamics to hydration layer dynamics contributing to the dynamical heterogeneities which merits further rigorous investigations. This will have strong implications on the dynamics of lipid rafts, skeleton fences to understand if the membrane organizations are driven by equilibrium processes. Moreover, it will be interesting to find out the length-scale dependence of dynamical heterogeneity using the block analysis of van Hove function and four point susceptibility. Importantly, if the information of length-scale of dynamic heterogeneities is embedded to the length-scale of the lipid phase transition, the analysis will allow in predicting a length-scale for the phase transition. Since confined biological water near membranes at room temperature exhibit similar dynamics as the super-cooled bulk water, this can enhance our understanding on the mechanisms of bioprotection during freezing stresses and have similar applications as on cryo-preservations, but at room temperature. At the same time, our findings raise few more questions: Is the coupling between lipid and hydration dynamics kinetic or thermodynamic in nature? Do interface water with large confinement lifetime have intermolecular energy flow via mode-coupling to keep the bonds intact? Is the nature of their motions cooperative?

V. ACKNOWLEDGEMENT

AD is thankful to the project IITJ/SEED/20140016 for financial support.

AD is thankful to the project IITJ/SEED/20140016 for financial support.

1Eisenber, D.; Kauzmann, W. The Structure and Properties of Water; Oxford University Press: New York: 1969.
2Jungwirth, P. The Journal of Physical Chemistry Letters 2015, 6, 2449–2451.
3Lingwood, D.; Simons, K. Science 2010, 327, 46–50.
4Muro, S. Cell 2003, 115, 377–388.
5He, W.; Song, H.; Su, Y.; Geng.; Ackerson, B. J.; Peng, H. B.; Tong, P. Nature Communications 2016, 7, 11701.
6Gowrishankar, K.; Ghosh, S.; Saha, S.; Rumamol, C.; Mayor, S.; Rao, M. Cell 2012, 149, 1353 – 1367.
7Mouritsen, O. G.; Jorgensen, K. BioEssays 1992, 14, 129–136.
8Roobala, C.; K. J. B. Soft Matter 2017, 13, 4598–4606.
9Debnath, A.; Ayappa, K. G.; Maiti, P. K. Physical Review Letters 2015, 115, 018303–1–5.
10Srivastava, A.; Debnath, A. The Journal of Chemical Physics 2018, 148, 094901.
11Ji, M.; Odelius, M.; Gaffney, K. J. Science 2010, 328, 1003–1005.
12Das, S.; Biswas, R.; Mukherjee, B. The Journal of Physical Chemistry B 2015, 119, 274–283.
13Balasubramanian, S.; Pal, S.; Bagchi, B. Physical Review Letters 2002, 89, 115505–1–4.
14Biswas, R.; Furtado, J.; Bagchi, B. The Journal of Chemical Physics 2013, 139, 144906–1–11.
15Pal, S. K.; Peon, J.; Bagchi, B.; Zewail, A. H. The Journal of Physical Chemistry B 2002, 106, 12376–12385.
16Xue, C.; Zheng, X.; Chen, K.; Tian, Y.; Hu, G. The Journal of Physical Chemistry Letters 2016, 7, 514–519.
17Pal, T.; Biswas, R. The Journal of Physical Chemistry B 2015, 119, 15683–15695.
18Indra, S.; Guchhait, B.; Biswas, R. The Journal of Chemical Physics 2016, 144, 124506.
19Bhattacharyya, S. M.; Bagchi, B.; Wolynes, P. G. Proceedings of the National Academy of Sciences 2008, 105, 16077–16082.
20Mallamace, F.; Branca, C.; Corsaro, C.; Leone, N.; Spooren, J.; Stanley, H. E.; Chen, S.-H. The Journal of Physical Chemistry B 2010, 114, 1870–1878.
21Gallo, P.; Rovere, M.; Spohr, E. The Journal of Chemical Physics 2000, 113, 11324–11335.
22Gallo, P.; Rovere, M.; Spohr, E. Physical Review Letters 2000, 85, 4317–4320.
23Bellissent-Funel, M.-C. Journal of Molecular Liquids 1998, 78, 19–28.
24Mostafa, Y.; J.-M., P. R.; Bilge, Y. Journal of the American Chemical Society 2011, 133, 2499–2510.
25Sciortino, F.; Gallo, P.; Tartaglia, P.; Chen, S. H. Physical Review E 1996, 54, 6331–6343.
26Gallo, P.; Sciortino, F.; Tartaglia, P.; Chen, S.-H. Physical Review Letter 1996, 76, 2730–2733.
27Mazza, M. G.; Giovanbattista, N.; Starr, F. W.; Stanley, H. E. Physical Review Letter 2006, 96, 057803.
28Jana, B.; Bagchi, B. The Journal of Physical Chemistry B 2009, 113, 2221–2224.
29Abascal, J. L. F.; Vega, C. The Journal of Chemical Physics 2005, 123, 234505–1–12.
30Debnath, A.; Mukherjee, B.; Ayappa, K. G.; Maiti, P. K.; Lin, S.-T. The Journal of Chemical Physics 2010, 133, 174704–1–14.
