1D07

Structure and orientation of water at charged and zwitterionic lipid/water interfaces

(Molecular Spectroscopy Laboratory, RIKEN)

o J. A. Mondal, S. Nihonyanagi, S. Yamaguchi, and T. Tahara

Interfacial water, which plays important roles in chemistry and biology, is less understood compared to the bulk water. Interaction between bio-molecules (e.g. lipids and proteins) and water is crucial for many biochemical processes and the structure and orientation of water are one of the most important factors for the diverse nature of interactions at bio-interfaces. We report the heterodyne-detected vibrational sum frequency generation (HD-VSFG) study of different lipid/water interfaces, which directly revealed net water orientation and hydrogen-bond strength at these biologically relevant interfaces.

To investigate interfacial water properties at the lipid/water interfaces, we used three different model lipids with varying headgroup structures. Figure 1 shows the chemical structures of these lipids; 1-palmitoyl-2-oleoyl-*sn*-glycero-3-phosphocholine (POPC), 1,2-dipalmitoyl-*sn*-glycero-3-phosphoglycerol (DPPG), and 1,2-dipalmitoyl-3-trimethylammonium propane (DPTAP), respectively. Hence forth these lipids were abbreviated by the acronym of their respective headgroups such as PC (POPC), PG (DPPG), and TA (DPTAP), respectively. Details of the HD-VSFG measurements were described elsewhere.[1]

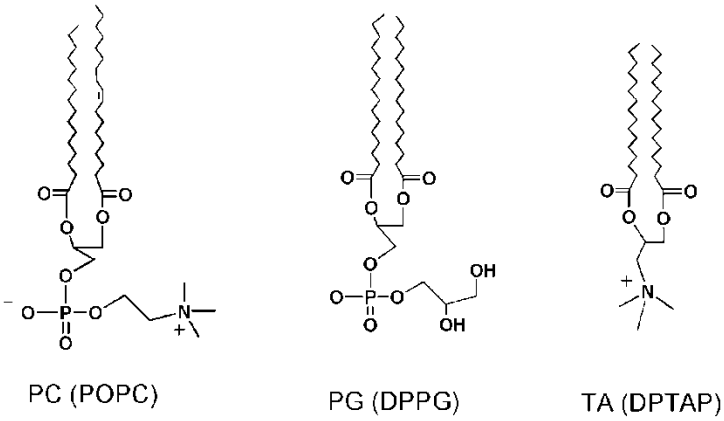

Figure 1. Chemical structures of three model lipids. PC is a zwitterionic (neutral) lipid. PG and TA are charged lipids.

Figure 2 shows the imaginary $\chi^{(2)}$ ($\mathrm{Im}\chi^{(2)}$; $\chi^{(2)}$ is the second-order nonlinear susceptibility) spectra of PC (black), PG (red), and TA (blue)/water interfaces in the OH stretch region. The $\mathrm{Im}\chi^{(2)}$ spectra of anionic (PG) and zwitterionic (PC) lipid/water interfaces have a positive sign, which indicates net hydrogen-up (H-up) orientation of interfacial water at these interfaces.[1] In the case of a cationic (TA) lipid/water interface, the $\mathrm{Im}\chi^{(2)}$ spectrum has a negative sign corresponding to the net hydrogen-down (H-down) orientation of interfacial water. For charged lipids (PG and TA), interfacial water orientation is governed by the net electric field at the interface.[2] In the case of the neutral lipid (zwitterionic; PC), there is no net electric field at the interface and, hence, the net water orientation at PC/water interface depends on the preferred orientation of water in the close vicinity of the phosphate and choline groups and on the net dipole moment at the interface.[3] In our knowledge, this is the first direct experimental evidence of different water orientations at various lipid/water interfaces.

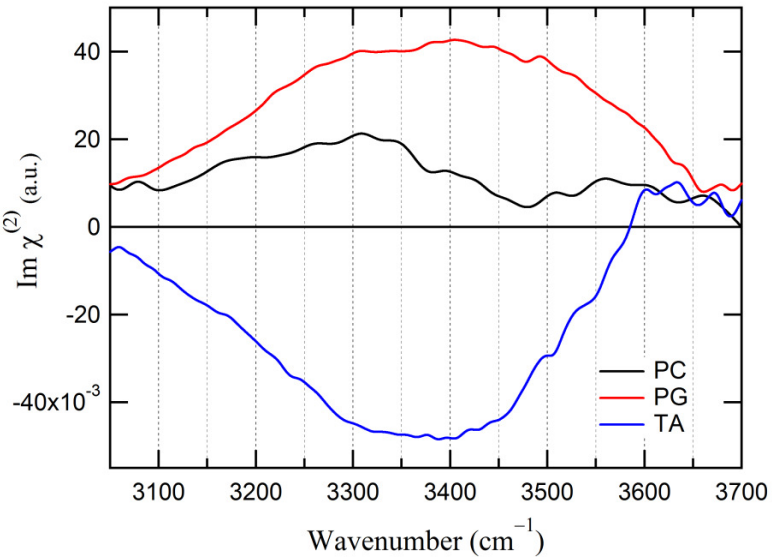

Figure 2. Im$\chi^{(2)}$ spectra of PG (red), TA (blue), and PC(black)/water interfaces in OH stretching region, measured with *ssp* polarization combination and surface pressure ≈ 25 ± 3 mN/m. Instead of neat water, isotopically diluted water (H_2O/HOD/D_2O = 1/8/16) was used.

Apart from the orientation of water at the lipid/water interfaces, another very important aspect is the structure of interfacial water. For the charged lipids (PG and TA), we observed a single broad band in the OH stretch region with maximum at ≈ 3400 cm^{-1}, as shown in Figure 2. This feature suggests that the interfacial water structure is similar to bulk water structure and there is no distinct "ice-like" structure.[4] Nevertheless, the maxima of the OH stretch bands in the Im$\chi^{(2)}$ spectra of PG and TA/water interfaces are very close to each other and similar to the maximum in the bulk HOD spectrum (≈ 3400 cm^{-1}). This manifests that the interfacial water has comparable hydrogen-bond strength at cationic and anionic lipid/water interfaces and it is analogous to that of bulk water on average.[2]

Unlike the charged lipids, the Im$\chi^{(2)}$ spectrum of PC/water interface has a positive band with a dip at ≈ 3470 cm^{-1} (black line, Figure 2). The red region of the positive band is predominantly due to the H-up oriented water molecules which are hydrogen-bonded with the negatively charged phosphate group.[5] The spectral dip with the minimum at ≈ 3470 cm^{-1} is originated from the H-down oriented water molecules associated with the choline group.[4] The water around the negatively charged phosphate group has strongly hydrogen-bonded water molecules and that around the positively charged choline group has relatively weakly hydrogen-bonded structure. Thus the Im$\chi^{(2)}$ spectrum of the PC/water interface directly manifests the presence of two distinct water structures around the phosphate and choline groups of zwitterionic PC. This is a demonstration of the preferred water orientation and distinct water structures at various lipid/water interfaces, which play a crucial role in many important biochemical reactions.

(1) Nihonyanagi, S.; Yamaguchi, S.; Tahara, T. *J. Chem. Phys.* **2009**, *130*, 204704.
(2) Mondal, J. A.; Nihonyanagi, S.; Yamaguchi, S.; Tahara, T. *J. Am. Chem. Soc.* **2010**, *in press*.
(3) Nagata, Y.; Mukamel, S. *J. Am. Chem. Soc.* **2010**, *132*, 6434-6442.
(4) Nihonyanagi, S.; Yamaguchi, S.; Tahara, T. *J. Am. Chem. Soc.* **2010**, *132*, 6867-6869.
(5) Zhao, W.; Moilanen, D. E.; Fenn, E. E.; Fayer, M. D. *J. Am. Chem. Soc.* **2008**, *130*, 13927-13937.