Effect of membrane fluidity change on long-range hydration dynamics observed by terahertz time domain spectroscopy

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***Abstract*—Composition of lipid membrane affects its fluidity and biological function. Here, we investigate the hydration extent and relaxation time of water around individual phospholipid bilayers with different fatty acids chain length using terahertz time domain spectroscopy (THz-TDS). This measurement suggests that long-range hydration state of lipids is associated with membrane fluidity. The detailed results will be presented in the conference.**

I. INTRODUCTION

Phospholipid bilayers, with membrane proteins, are main components of cell membrane to control the movement of materials into and out of cells. The fact that maintaining appropriate mobility of membrane components is critical to carrying out this role has inspired many researchers to study on membrane fluidity. The factors determining fluidity of lipid bilayers found in studies of past decades include temperature, length and saturation of fatty acids chain, cholesterol concentration, etc [1]. Because frequency range of molecular motions of membrane lipids are MHz-GHz, these results have been mainly obtained from EPR [2], NMR[3], Fluorescence spectroscopy [4].

Especially in recent 10 years, a series of studies to associate hydration state of lipid bilayers with membrane fluidity have been done using Time Dependent Fluorescence Shift (TDFS) method [5]. However, TDFS method has limitations observing only a localized area around the fluorescent dye which is located in the bilayer inner space and indirectly predicting lipid mobility through the state of tightly bound hydration water in polar head interfacial region or hydrophobic core.

Recently, novel experimental techniques available for monitoring the ultrafast time scale (~THz) dynamics of water molecules have emerged; terahertz time domain spectroscopy (THz-TDS) [6,7] and pump-probe spectroscopy [6,8]. Consequently, the long-range hydration effect of lipid bilayers, i.e. the presence of weakly perturbed water molecules predicted by molecular dynamics simulations [9] has been confirmed [10]. This result has given a new clue to the membrane research directions and techniques toward the relationship between hydration water dynamics and lipid membrane properties. With this as a momentum, Choi *et al.* have reported with THz-TDS that drastic changes appear in long-range hydration water dynamics above the lipid phase transition temperature [11].

In the current study, varying the fatty acids chain length among some factors that determine membrane fluidity, the hydration degree of lipid bilayers is measured by using attenuated total reflection THz-TDS. Comparing these result with the previous studies, the relationship between lipid fluidity and long-range hydration state is demonstrated.

II. RESULTS

Several kinds of phosphatidylcholines (PCs) with different fatty acids chain length (12:0 DLPC, 14:0 DMPC and 16:0 DPPC) are purchased from Avanti Polar Lipids, Inc. in the powder form and dissolved in de-ionized water (MilliQ). The longer the fatty acids chain length, the higher the transition temperature of the phospholipid. Figure 1 shows phase transition temperature depending on the acyl chain length of phosphatidylcholines.

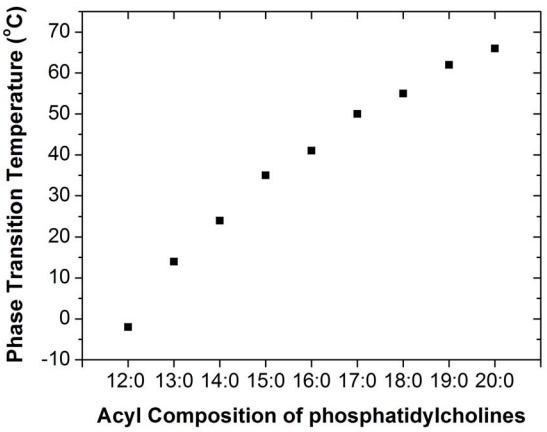

Fig. 1. Phase transition temperature for saturated phosphatidylcholines. Transition temperature data was obtained from Avanti Polar Lipids, Inc. [12].

The lipid suspensions are maintained above phase transition temperature during the hydration period of 1 hour with vigorous shaking to hydrate adequately in its fluid phase and form the multi-lamellar vesicles. All samples have same lipid concentration (the molar ratio: $R = [H_2O]/[PC] = 250$, above the fully hydrated condition and the critical micelle concentration [12]) and are maintained at a constant temperature of 45±0.1°C (in fluid phase) during all measurements.

Experiments are carried out by THz-TDS attenuated total reflection module (TAS7500SP, Advantest Corporation) for the accurate determination of the complex dielectric constant of the solutions. Figure 2 shows the imaginary part of the dielectric constant of pure water, 12:0 DLPC, 14:0 DMPC and 16:0 DPPC solutions in the THz region. Due to the weak absorption by the PC molecules and non-dispersive dielectric response in this region, the complex dielectric constant of PC solutions mainly comes from the dynamics of water molecules [10]. The slow and fast relaxation of water dipole moment is dominant at THz frequency region studied here (0.5–1.5 THz) [13].

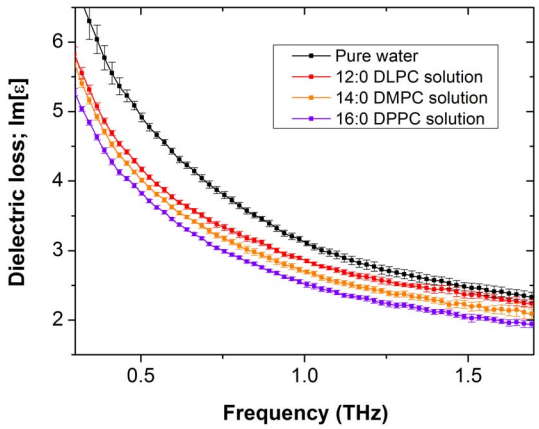

Fig. 2. The imaginary part of dielectric constant of pure water, 12:0 DLPC, 14:0 DMPC and 16:0 DPPC solutions, obtained by THz TDS measurements. The solid lines are the guides to the eyes.

The longer the fatty acids chain length, the larger the reduction of the dielectric constant of PC solution from that of pure water is observed here. The additional results and detailed analysis will be presented in the conference.

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