Spatiotemporal features of nucleic acid hydration and their changes during denaturation revealed by THz spectroscopy

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***Abstract*— Dynamics and structure of water network around nucleic acid have been subjects of intensive studies due to their contributions to biological process. Here, we identified spatiotemporal features of DNA hydration through examining fully-hydrated DNA solutions by THz spectroscopy. The results suggest the presence of weakly bound water beyond tight-binding hydration layer with the timescale of 11.6ps and the thickness of 7.6Å. Also, we will present changes in the hydration water during DNA denaturation at the conference.**

I. INTRODUCTION

Primary function of DNA is transferring genetic information to RNA or another DNA accurately. This function is associated with a conformational change of DNA from double helix to single strand, so-called DNA denaturation. However, it is reported that considerable amount of water molecules interacts with double helical DNA [1, 2]. Studies on changes in structure and dynamics of surrounding water caused by denaturation have attracted researcher's attentions, as the surrounding water is critical to stabilizing double helix structure [3, 4]. Thus far, behaviors of these water molecules upon DNA denaturation have been examined by various techniques such as NMR, dielectric measurement and neutron scattering [3-5].

Development of THz spectroscopy enables scientists to observe picosecond-dynamics of water directly [6-7]. Conventional techniques measured relatively slow water motions (up to 100ps) such that they examined tightly bound water adjacent to DNA. However, recently, more flexible DNA-bound water is observed by fluorescence spectroscopy and expected to play a role in recognition process due to its dynamic nature [8].

Although several studies have been done to understand structural and dynamical properties of surrounding water upon DNA denaturation, a relationship between behavior of water and DNA denaturation is not clear yet. In this paper, we determine the extent of DNA-bound water in the range of picosecond timescale and its changes during DNA denaturation using THz spectroscopy.

II. RESULTS

A twelve base-paired duplex is dissolved in buffer and aqueous solutions at several DNA concentrations are examined by THz time-domain spectroscopy. We assumed that measured dielectric spectra of DNA solutions mostly originate in water because dielectric response of dehydrated DNA film is low-loss and dispersionless (Fig. 1).

Both dielectric constants and volume fraction of DNA molecule with hydration water are extracted using following Bruggeman effective medium theory.

$$v_h \frac{\varepsilon_h^* - \varepsilon_{sol}^*}{\varepsilon_h^* + 2\varepsilon_{sol}^*} + (1 - v_h) \frac{\varepsilon_b^* - \varepsilon_{sol}^*}{\varepsilon_b^* + 2\varepsilon_{sol}^*} = 0$$

Here, v_h is a volume fraction of hydration water plus DNA molecule. ε^*_h, ε^*_{sol}, and ε^*_b are complex dielectric constants of hydration water plus DNA, DNA solution and buffer, respectively.

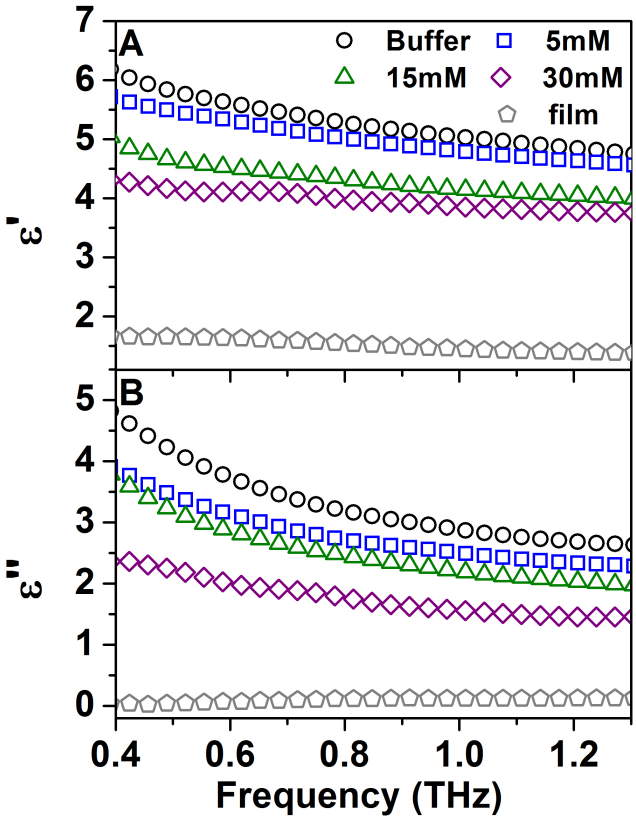

Fig 1. Dielectric responses of several DNA solutions and DNA film

When fitting data with the equation above, the dielectric constant of hydration water plus DNA (ε^*_h) is assumed to be constant with respect to DNA concentration, while the volume fraction (v_h) changes with the concentration. We obtained both timescale of reorientational motion and thickness of DNA hydration water from the calculated the dielectric constant and the volume fraction of hydration water plus DNA.

Relaxation time of reorientational motion of DNA hydration water is obtained to be 11.6ps from Double Debye model. This timescale is much faster than nanosecond-timescale of tightly bound water observed by NMR or dielectric measurement but still slower than 7ps of bulk water. Therefore, 11.6ps of reorientation time demonstrates the presence of weakly bound water around DNA. From the volume fraction of DNA plus hydration water, the average radius of DNA with hydration water is calculated to be 24.4Å. When considering a radius of 16.8Å acquired from dielectric measurement which includes tightly bound water, the thickness of weak-binding hydration water is 7.6Å, which corresponds to 57 water molecules per nucleotide (Fig. 2). The result suggests that DNA molecule has longer influence on water network than previously observed.

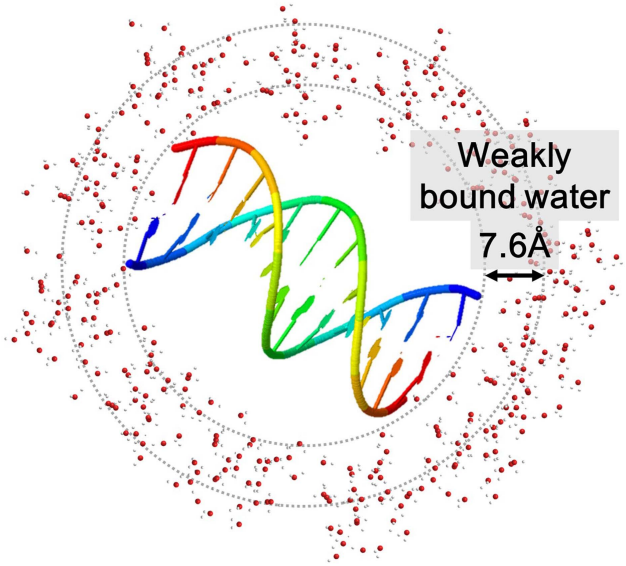

Fig 2. Schematic diagram for hydration layer of DNA

In order to study changes in this weakly bound water upon DNA denaturation, we confirmed structural transition from double stranded DNA (dsDNA) to single stranded DNA (ssDNA) during denaturation using circular dichroism (CD) spectroscopy (Fig. 3). As temperature increases, signatures of B conformation with a negative peak at ~245nm and a positive peak at ~275nm apparently disappear. The degree of DNA denaturation is measured by UV-Visible spectroscopy and spatiotemporal changes in weakly bound water of DNA are examined using THz spectroscopy. Details will be shown at the conference.

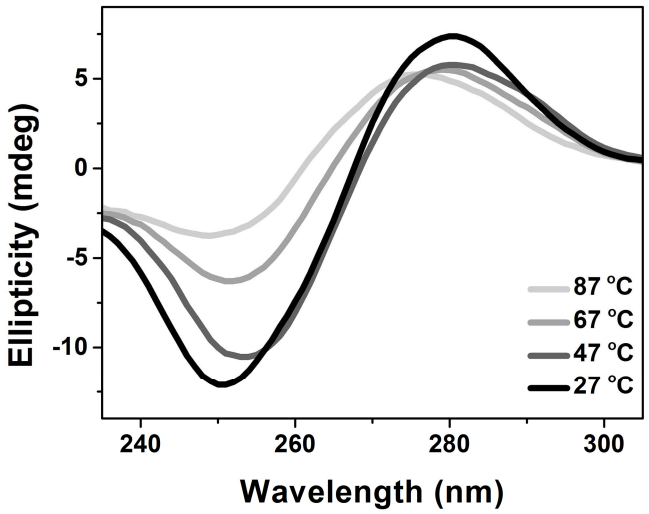

Fig 3. CD spectra of DNA solutions at temperature from 27 ℃ to 87℃

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