Molecular dependence of collagen model peptide stuidied by low frequency Raman spectroscopy

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Introduction: Low frequency Raman spectra reflects the intermolecular vibration and the interactions between molecular chains. Especially, low frequency Raman spectroscopy is a powerful tool for monitoring higher-order structures of polymer in water. Therefore, we tried to investigate the structural changes of collagen model peptide by low frequency Raman spectroscopy.

Experimental Section: Two kinds of collagen model peptides ((Pro-Pro-Gly)$_{10}$ (PPG10) and (Pro-Pro-Gly)$_5$ (PPG5)) are used as samples (Figure 1). PPG10 and PPG5 are dissolved in D_2O. HR-800-LWR (HORIBA) and SureBlock XLF (Ondax) are used for this study. Each range of measurements are from 70 cm^{-1} to 3200 cm^{-1} and from 20 cm^{-1} to 1590 cm^{-1}. T95-HS (LINKAM) is used as a temperature controller.

Results and Discussion: PPG10 forms a triple-helical structure like collagen in aqueous solution, and denatures when it is heated. This structural change from a triple-helix to a random coil in aqueous solution occurs reversibility in PPG10[1]. However, PPG5 does not show such a conformational change[2]. Figure 2 shows temperature dependent low frequency Raman spectra and their second derivative of PPG10/D_2O and PPG5/D_2O from 27.8 ^oC to 63.2 ^oC. They were given by denoising by singular value decomposition, curve fitting, normalization, and subtraction $(\text{each data} - 70\ ^oC)$. There are three peaks appeared at around 259 cm^{-1}, 178 cm^{-1}, and 106 cm^{-1} in the 80-300 cm^{-1} region. Figure 3 shows the change of the peak position at around 178 cm^{-1} and 106 cm^{-1}. During cooling process, the peak at around 178 cm^{-1} of PPG10 showed a shift to lower frequency region. While that of PPG5 did not change with temperature (Figure 3 (a)). Moreover, the peak at around 106 cm^{-1} of PPG10 shifted to the higher frequency region, while that of PPG5 did not change with temperature (Figure 3 (b)). It is already reported that the peak at 178 cm^{-1} due to the water and 106 cm^{-1} reflected to the intermolecular hydrogen bond in the amide group[3,4]. Therefore, the

lower frequency shift of the peak at 178 cm^{-1} due to water indicated that the hydrogen bond between water-to-water is disturbed by interaction between collagen model peptide and water when temperature becomes higher than the coil-helix transition temperature, but it is not interfered when temperature becomes lower than the temperature which is the formation of triple-helical structure. And the higher frequency shift of 106 cm^{-1} indicated that the intermolecular hydrogen bond in the amide group of the collagen model peptide is stronger because triple-helical structure is formed. These results show that the triple-helical structure of collagen model peptide are formed between 40 to 50 ^{o}C. We were able to demonstrate that structural changes from helix to random coil by low frequency Raman spectroscopy.

References:

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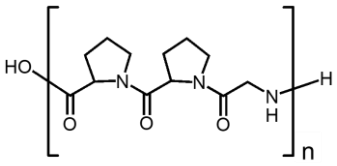

Figure 1 Chemical structure of PPG10 (n= 10) and PPG5 (n= 5)

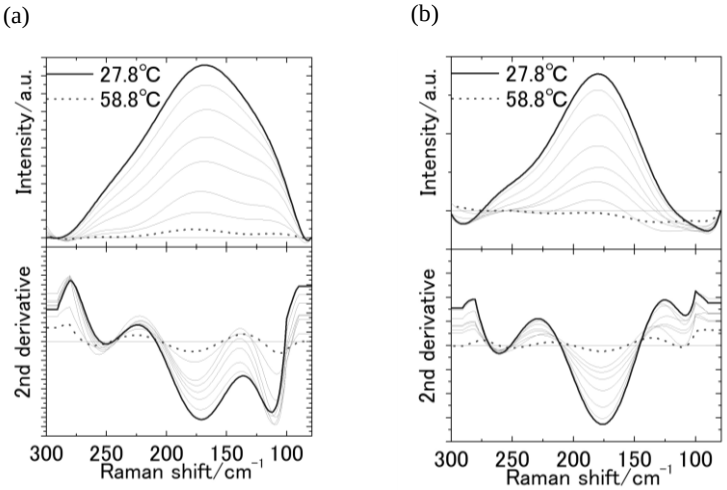

Figure 2 The Raman spectra and their 2nd derivative spectra of (a) PPG10 (b) PPG5

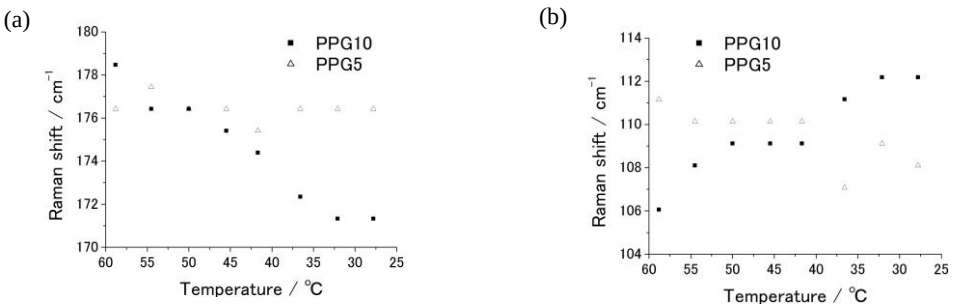

Figure 3 The peak position of PPG10 and PPG5 at around (a) 178 cm^{-1} (b) 106 cm^{-1}