Infrared Spectroscopic Study of Molecular Fastening by Mechanical Compression in an Elastic Film

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An amphiphilic compound that has a consecutive hexaleucine array (Leu-6) was designed to have molecular interaction via hydrogen bonding between the hexaleucinate moieties, and also have molecular interdigitation mechanism named "leucine fastener" at the same moieties, which is expected to help the molecular network formation more rigid. In the present study, the leucine fastener has been obtained by a simple hydraulic press of a bulk Leu-6 solid. The elastic film thus obtained exhibited a characteristic infrared marker band at 2985 $\mathrm{cm^{-1}}$, which has proved the leucine fastener formation.

Molecular assemblies based on a noncovalent-bond intermolecular linkage have a benefit that characteristics of the materials can be controlled via molecular design of the material architecture. The materials of the noncovalently organized molecular assemblies are now recognized to be a member of polymer, which is often enhanced as "supramolecular polymer" as proposed by Meijer et al.[1]

Yamada et al.[2] recently added another concept to the supramolecular polymer by introducing a new molecular interaction named "leucine fastener" that is a molecular interdigitation between the side chains of leucinates within β-sheets. The new noncovalent-bond linkage works powerfully to have the molecular assemblies in a monolayer highly ordered. The compound consists of an amino acid moiety including n-leucine array ($n = 3$ and 4) and hydrocarbon tails, which is denoted as Leu-n (Chart 1). The compounds form a stable Langmuir monolayer at the air–water interface. The reason that leucine was chosen as an amino acid is that the Y-shaped leucine residues work as gear teeth of molecular fastening. The amino-acid array has another role that it forms β-sheets via intermolecular hydrogen bonding whose direction is perpendicular to that of the leucine fastener (Chart 1).[3] In this manner, Leu-n was designed to have a two-dimensional molecular assembly in a monolayer, which was readily proved by infrared multiple-angle incidence resolution spectrometric (MAIRS)[4,5] and atomic force microscopic (AFM) studies of Langmuir–Blodgett (LB) films.

The unique molecular assemblies can also be prepared in a cast film on a hydrophobic surface when $n = 3$ and 4, and the removed film from the surface exhibits an extraordinarily strong elasticity for the noncovalently assembled molecular film.[5] In both LB and cast films, the formation of the leucine fastener was commonly found to have a relation to appearance of an infrared absorption band at about 2985 $\mathrm{cm^{-1}}$,[5] although the assignment of the band was not confirmed. A film of the compound with a leucine array of $n < 4$ prepared by a hydraulic press, however, did not exhibit elasticity. It was considered that the leucine array of $n < 4$ was too short to form the leucine fastener by the mechanical press only.

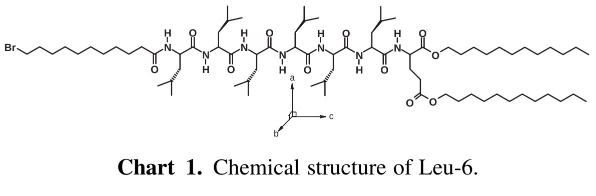

Chart 1. Chemical structure of Leu-6.

In the present study, a similar compound with a longer leucine array (Leu-6) has been synthesized for studying its molecular interdigitation. The longer array was expected to exhibit the leucine fastener under a simple hydraulic press.

Details of the synthesis of $\mathrm{Br{-}(CH_2)_{10}CO{-}Leu_6{-}Glu{-}(OC_{12}H_{25})_2}$ (Leu-6) are described elsewhere.[6] Leu-6 was hard to be dissolved in many solvents, and a chloroform solution after sonication became a gel. A xerogel of Leu-6 was therefore obtained by addition of methanol (poor solvent) into a chloroform gel of Leu-6 followed by air drying.

Before pressing Leu-6, a cast film of Leu-6 was prepared by spreading the gel on a hydrophobic surface and the cast film was removed after the evaporation of the solvent. The cast film was found to be brittle and no elasticity was found. Since the cast films of Leu-3 and -4 had an apparent elastic property, the drastic change should be attributed to the difference of the leucine array in length. In this manner, the large difference of the elastic property strongly suggests that molecular interaction of Leu-6 is largely different from those of Leu-3 and -4. The poor elasticity of Leu-6 suggests that the molecular interdigitation of the leucine arrays is difficult to happen, probably because the length of the array is too long for spontaneous interdigitation. If this speculation is true, an external pressure would help the interdigitation.

Then, Leu-6 was subjected to a hydraulic press that was performed at a high pressure of 10^2–10^3 $\mathrm{kg\,cm^{-2}}$. As a result, the pressed Leu-6 film has been found to have elasticity like the cast films of Leu-3 and -4. This clearly indicates that the external pressure played an important role to make the molecules be interdigitated as expected. To investigate the molecular interdigitation in more detail, three samples of Leu-6 in different states were measured by infrared attenuated total reflection (ATR) spectrometry.

The cast film of Leu-6 for the infrared ATR measurements was prepared by spreading a chloroform gel of Leu-6 (ca. 0.3 $\mathrm{g\,L^{-1}}$) of 50 μL on the flat surface of a semispherical ATR prism made of germanium (Ge). The prism was purchased from Pier Optics (Gunma, Japan). Infrared ATR spectra of the Leu-6

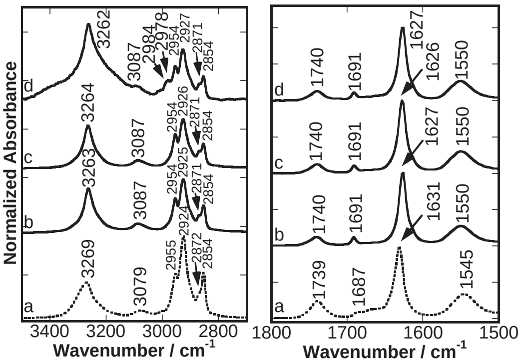

Figure 1. IR ATR spectra of (a) a Leu-3 cast film (dotted line), (b) a Leu-6 cast film, (c) a xerogel of Leu-6, and (d) a hydraulically pressed film of Leu-6.

cast film were measured with an unpolarized infrared ray at an angle of incidence of 50° from the surface normal.

An infrared ATR spectrum of the Leu-6 cast film deposited on Ge is presented in Figure 1b. The amide I band that is mainly attributed to the C=O stretching vibration (νC=O) mode is found to be split into two bands at 1691 and 1627 cm^{-1}. These band locations indicate that the "anti-parallel" β-sheet structure is formed in the film as a result of intermolecular hydrogen bonding between C=O and N–H.[2,5] This is supported by the sharp amide A band at 3263 cm^{-1}, which is mainly attributed to the N–H stretching vibration (νN–H) mode. This result is striking because the amide bands that are specific to the "parallel" β-sheet were found for compounds with a shorter leucine array. For example, a Leu-3 cast film prepared on CaF_2 gives the amide I band as a singlet peak at 1633 cm^{-1} (data not shown),[2] which clearly indicates that there is no anti-parallel β-sheet in the film. Another related spectrum is presented in Figure 1a by a dotted line, which is an infrared ATR spectrum of a Leu-"3" cast film on the ATR prism surface of Ge. In the spectrum, the amide I band appears at 1631 cm^{-1} accompanied by a minor band at 1687 cm^{-1}. The band locations and the intensity tell us that the Leu-3 cast film comprises both the parallel and anti-parallel β-sheets. On the other hand, a Leu-4 cast film was reported to give the amide I band at 1628 cm^{-1} with a clearer sub-peak at 1688 cm^{-1}, which means that the anti-parallel β-sheet is a major component in the Leu-4 cast film. In this manner, the amide bands of the Leu-6 (Figure 1b) strongly suggest that a Leu compound with a longer leucine array has a higher content of the anti-parallel β-sheet.

Besides, the CH_2 symmetric and anti-symmetric (ν_sCH_2 and ν_aCH_2) vibration bands appear at 2854 and 2925 cm^{-1}, respectively, both of which imply that the hydrocarbon chain has the gauche conformation, i.e., disordered chains. The disordered chains were also found in the compounds with a shorter leucine array irrespective of sampling techniques. In the case of a Leu-6 cast film, the brittle character reflects low crystallinity along with the disordered conformation. According to Yamada et al.,[2] the low crystallinity is inevitable for realizing an elastic material made of noncovalent-bond linkage, which is called "hierarchically integrated supramolecular polymer." The low crystallinity of Leu-6 satisfies one of the inevitable factors, but another factor is necessary to make the brittle character to elastic.

For reference, an infrared ATR spectrum of the xerogel was measured as presented in Figure 1c. The spectrum pattern is almost identical to that of the cast film, which implies that crystallization or molecular rearrangement is ignorable in the cast film. In other words, the influence of the substrate surface on the cast film structure is ignorable for Leu-6.

Then, a pressed Leu-6 film with a good elasticity obtained by a hydraulic press was subjected to infrared ATR spectrometry. Therefore, the large difference should be attributed to the sample preparation technique, and the hydraulic press is considered to play a role of another "inevitable factor" to make the material elastic. If the elastic property reflects the leucine fastener, it should also be reflected in the spectrum. The measured spectrum is presented in Figure 1d.

The fundamental characteristics of the spectrum in the low wavenumber region (1800–1500 cm^{-1}) are similar to those of the cast film and the xerogel. Therefore, the anti-parallel β-sheet structure seems retained even after applying a high pressure. Regardless, a large broad band appears being overlaid on the amide A band at 3262 cm^{-1}, which is obviously different from other spectra (Figures 1b and 1c). The band broadening is considered to reflect a wide variation of hydrogen bonding, which means that the anti-parallel β-sheets are a little damaged as a result of the hydraulic press, since a similar damage was also found in a monolayer of Leu-4 under a high surface pressure.[5] In this manner, the pressed film of Leu-6 has a similar characteristic to that of the monolayer of Leu-4. In other words, the two samples can be compared to discuss their molecular interaction.

Of another importance is appearance of new bands at 2984 and 2978 cm^{-1} in the spectrum of the pressed film (Figure 1d). One of these bands was already recognized in the infrared MAIRS spectra of a LB film of Leu-4,[5] in which the band at 2985 cm^{-1} appeared as a shoulder. A common principle of the pressed and LB films is the molecular interdigitation via the leucine fastener. Therefore, the band at ca. 2985 cm^{-1} is concluded to be a marker band of the leucine fastener. The other band at 2978 cm^{-1} suggests that an inferior leucine fastening portion coexists in the hydraulically pressed film to that in the monolayer.

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