Chirality Control of Self-Assembling Organogels from a Lipophilic L-Glutamide Derivative with Metal Chlorides†

Hirotaka Ihara,*,‡ Toshihiko Sakurai,‡ Taisuke Yamada,‡ Toshitaka Hashimoto,‡ Makoto Takafuji,‡ Takashi Sagawa,§ and Hiroshi Hachisako||

Department of Applied Chemistry & Biochemistry, Kumamoto University, Kumamoto 860-8555, Japan, Institute of Advanced Energy, Kyoto University, Uji 611-0011, Japan, and Department of Applied Life Science, Sojo University, Kumamoto 860-0082, Japan

Received January 15, 2002. In Final Form: March 6, 2002

A double-chain alkyl lipid with an isoquinoline-containing headgroup was derived from L-glutamide. This worked as an organogelator in benzene, cyclohexane, a cyclohexanes−ethanol mixture, and so forth. Transmission electron microscope observation showed that the gelation was brought about through the formation of a fibrillar network, and circular dichroism (CD) spectra indicate that the fibrillar aggregates were based on highly ordered structures. The critical aggregation concentration (cac) was estimated by detecting the enhancement of CD strength due to molecular orientation. The cac's were dependent on solvents and always lower than the critical gelation concentration. This indicates that the sol state includes supramolecular aggregates inside which the lipids are well developed. In addition, it was confirmed that the chirality and morphology of the aggregates are remarkably perturbed by chelating with metal chlorides. In the case of $CuCl_2$ which can form a square planar coordination, the chirality enhancement was observed, but in the case of $CoCl_2$ and $ZnCl_2$ which can form an octahedral coordination state, serious morphological change was observed with remarkable decrease of the chirality.

Introduction

It is known that special synthetic lipids work as self-assembling organogelators.[1−3] These gelations have often been brought about through network formation with well-developed fibrous aggregates of their lipids. To make organogels, many synthetic lipids have been derived from peptides[4−13] and sugars[14−16] because the hydrophobic effect is available only in aqueous systems and thus direct interaction such as hydrogen bonding interaction is essentially useful for molecular aggregation in organic solvents. Also, the chirality of lipids plays important roles in the formation of fibrillar aggregates. Therefore, the organogels often include suprastructural aggregates[4−20] such as helical and twisted ribbonlike aggregates with specific chirality. These unique features should open up possible applications as molecular materials for nanodevicing and chiral organic media for organic synthesis and chiral separation although few applications have been yet reported.[4] It should be very important to establish a method to control the chirality of organogels, and thus it promises to expand the possibilities. For this purpose, we newly synthesized an isoquinoline-containing lipid **1**. This can be characterized by the L-glutamide moiety (L-Glu) as a chiral source, double long-chain alkyl groups as lipophilic parts, and a headgroup (X) for chelation with metal ions. Also, the L-glutamide moiety with three amide bondings is mostly important to produce nanofibrous aggregates based on highly ordered structures such as lipid bilayer membranes. The typical examples have been reported as shown by lipids **2**−**6** (Figure 1).[4−9,21−28]

* Corresponding author. Telephone: +81-96-342-3661. Fax: +81-96-342-3662. E-mail: ihara@chem.kumamoto-u.ac.jp.

† This article is part of the special issue of *Langmuir* devoted to the emerging field of self-assembled fibrillar networks.

‡ Kumamoto University.

§ Kyoto University.

|| Sojo University.

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10.1021/la025535f CCC: $22.00
Published on Web 06/06/2002

Table 1. Apparent Dispersion States of 1-Containing Solutions

	ethanol	chloroform	benzene	toluene	cyclohexane	cyclohexane[a]
at 70 °C	clear sol	clear sol	clear sol	clear sol	clear sol	clear sol
at 10 °C	precipitated	clear sol	clear gel	clear gel	slightly turbid gel	clear gel

[a] Cyclohexane−ethanol = 100:1.

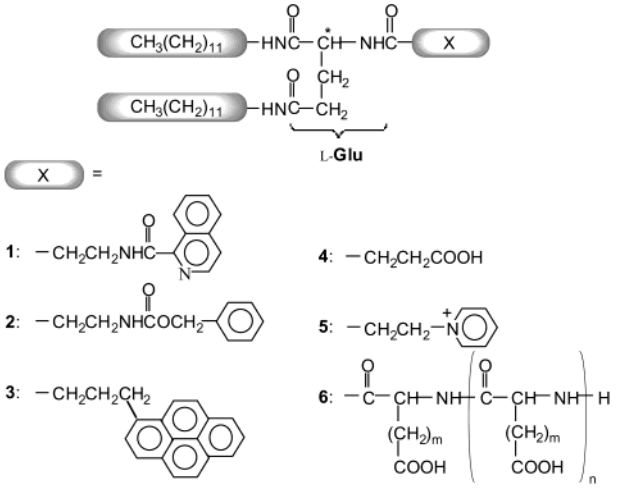

Figure 1. The chemical structures of L-glutamic acid derived lipids.

In this paper, we report that an L-glutamic acid derived lipid **1** forms self-assembling organogels and that the chirality can be controlled by recognizing the geometrical difference of the coordination state of metal ions, and the microenvironmental change is accompanied by macroscopic change.

Experimental Section

Materials. Lipid **2** was prepared according to our method reported previously.[4,8,27] Lipid **1** was derived through lipid **2** as follows: **2** was dissolved in hot ethanol and then Pd (5%) on carbon was added to the solution. When hydrogen gas was bubbled at 60 °C for 10 h, the debenzyloxycarbonylate was obtained. After recrystallization from ethanol, this was mixed with 1-isoquinoline in tetrahydrofuran. Diethyl phosphorocyanidate as a coupling reagent and triethylamine as a promoter were added to the solution and then stirred at 40 °C for 24 h. After the solvent was removed in vacuo, the residue was dissolved in chloroform and washed with a 0.2 N NaOH aqueous solution and water. **1** was obtained after recrystallization from methanol: mp = 143−146 °C.

UV and CD Measurements. The lipid **1** was dispersed in organic solvents at room temperature and then heated to be dissolved at 70−80 °C. The solution was rapidly added into a 1-mm quartz cell which was put into the cell holder (maintained at 70 °C) of a Hitachi U-2000 spectrophotometer. After the temperature was reduced stepwise at 70, 60, 50, 40, 30, 20, 10, and 5 °C, the UV spectral measurement was carried out after incubation for at least 20 min at these temperatures. According to a similar procedure, circular dichroism (CD) spectra were measured with a JASCO J-725 spectropolarimeter.

Metal chloride containing solutions were prepared as follows: a given amount of **1** was dissolved into 25 mL of a hot cyclohexane. An ethanol solution of metal chloride was added to the **1** solution, and then the content of ethanol was adjusted to be 1.0% v/v.

Inverse Fluid Method. The critical gelation concentration (cgc) and temperature were determined by a usual inverse fluid method using a ϕ 14 mm sample tube thermostated at each temperature.

Differential Scanning Calorimetry (DSC). DSC was carried out using EXTRA 6000 with DSC6200 from Seiko Instruments Inc. The thermograms were obtained using 80 mM of the sample and using a cooling rate of 1 °C.

Transmission Electron Microscopy (TEM). Transmission electron micrographs were recorded by using a JEOL 2000FX. The samples were spotted on carbon-coated copper grids (200 A). After excess of the samples was removed by a tissue paper and air-dried, they were stained with 2 wt % of molybdate.

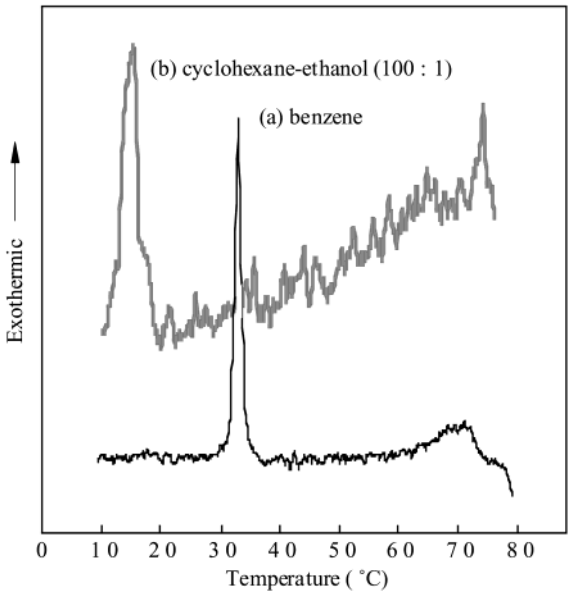

Figure 2. DSC thermograms of **1** (80 mM) in the cooling process.

Results and Discussion

Organogel Formation. Lipid **1** (5 mM) was readily dissolved in hot benzene, and then the solution was allowed to stand at 10 °C to make a clear gel. Similar gelation was also observed in toluene, cyclohexane, ethanol, and a cyclohexanes−ethanol mixture as shown in Table 1.

It was observed visually that the sol−gel transition in the cooling process occurred at around 35−40 and 15−20 °C in benzene and a cyclohexanes−ethanol (100:1) mixture, respectively. These transitions can be observed by DSC.[4,8,29,30] The DSC measurement showed that thermally induced sol-to-gel transitions were detected with exothermic peaks whose peak-top temperatures were located at 33 and 15 °C in benzene and a cyclohexanes−ethanol (100:1) mixture, respectively, as shown in Figure 2.

Lipid **1** also showed a lyotropic phase transition between sol and gel states. The cgc was dependent on a kind of solvent and estimated by an inverse fluid method to be 1−5 and 0.5−1 mM at 10 °C in the case of benzene and a cyclohexanes−ethanol (100:1) mixture, respectively. TEM observations indicated that the gels included well-developed fibrous aggregates as shown in Figure 3. This has been commonly found in organogels from low molecular weight compounds,[1] and thus the gelation was

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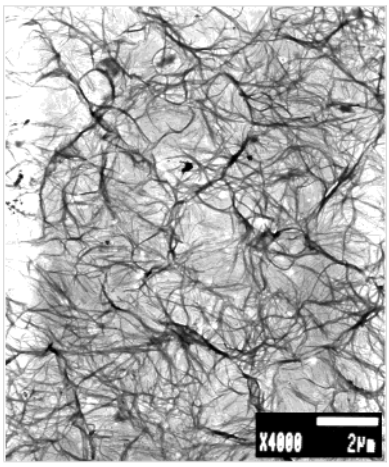

Figure 3. Transmission electron micrograph of aggregates of **1** in cyclohexane−ethanol (100:1). Initial concentration of **1** = 0.3 mM.

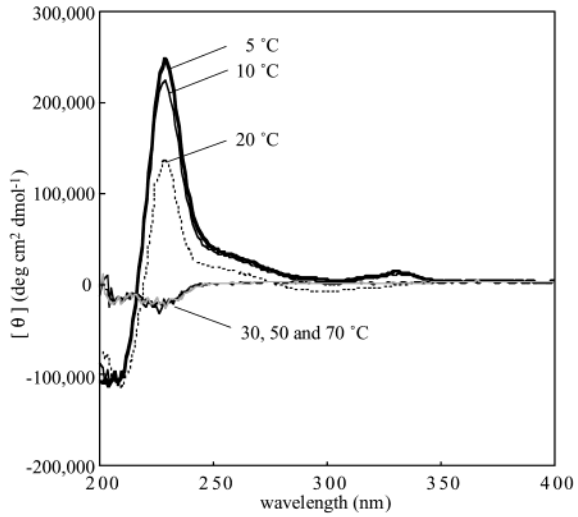

Figure 4. Temperature dependency of CD spectra with 0.3 mM **1** in a cyclohexane−ethanol (100:1) solution.

brought about through network formation of fibrous aggregates.

Critical Aggregation Concentration. Macroscopic measurements were not always helpful to understand microscopic aggregation states. For example, the value of cgc is absolutely dependent on the viscosity of solution and the interconnection density and strength of fibrillar aggregates. TEM observation also may not provide true aggregation morphology because the concentration process is necessarily included. Therefore, we adopted CD spectral measurement to evaluate the aggregation state. This method has been often used for aqueous membrane systems from chiral lipids with chromophoric groups.[23,24,31,32] Figure 4 showed that special CD patterns were obtained for a 0.3 mM solution of **1** at 5−20 °C but not at 30−70 °C. This was also accompanied by temperature-dependent blue-shifts at around 230 and 325 nm in the UV spectra. These phenomena can be explained by formation of supramolecular aggregates with chiral arrangement or chiral stacking among the isoquinoline moiety.

Effect of Metal Chlorides on Chirality. The most significant purpose of this work is to control the chirality

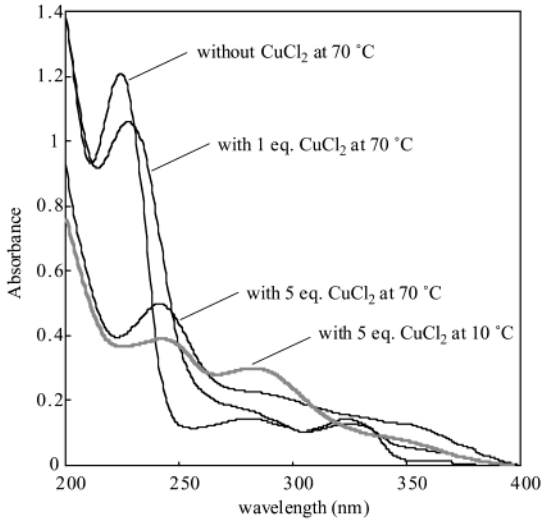

Figure 5. Temperature dependency of UV spectra with **1** (0.3 mM) and $CuCl_2$ in cyclohexane−ethanol (100:1) solutions.

of the gels by chelating with metal ions. For this purpose, we introduced an isoquinoline moiety as a chelating headgroup into the lipid, while some researchers have adopted crown ethers,[33,34] D-glucose derivatives,[35] and bipyridine derivatives,[36] and the chelating with metal ions was investigated. An isoquinoline moiety gives us some advantages: (1) the compact and planar structures do not interfere with the molecular orientation among the lipids and (2) it is usable as a sensitive chromophoric group to evaluate physicochemical properties of the aggregates.

In this study, we focused on Cu^{2+} and Co^{2+} with different coordination structures. $CuCl_2$ was very hard to dissolve in cyclohexane. When it was added as an ethanol solution to cyclohexane, almost no increase was observed at the absorption band of $CuCl_2$ because of precipitation. However, in the case of the **1**-containing cylohexane solution, it was easy to observe distinct spectral change as shown in Figure 5. This result indicates that the isoquinoline moiety is influenced by $CuCl_2$ and thus increases the solubility of $CuCl_2$ into cyclohexane. This came under other metal chlorides such as $CoCl_2$ and $ZnCl_2$. On the other hand, if **1** was replaced into **2**, the UV spectrum did not show any change for addition of these metal chlorides. It has been already described in detail that **2** forms self-assembling organogels.[4−6] The explanation for this is that **2** has a similar chemical structure to **1** but does not have any effective interaction source such as an isoquinoline moiety.

The effect of metal chlorides on the chirality was mainly investigated using a 0.3 mM solution of **1** in a cyclohexanes−ethanol (100:1) mixture. Figure 6 shows that the action of $CuCl_2$ is absolutely different from that of $CoCl_2$. In the case of $CuCl_2$, distinct CD spectral change was induced with new cotton effects at 245, 260, and 295 nm at 10 °C (Figure 6a) and a similar chirality was also observed at 70 °C (Figure 6b). On the other hand, in the case of $CoCl_2$, the original chirality almost disappeared at 10 °C (Figure 6c) and almost no chirality was observed at

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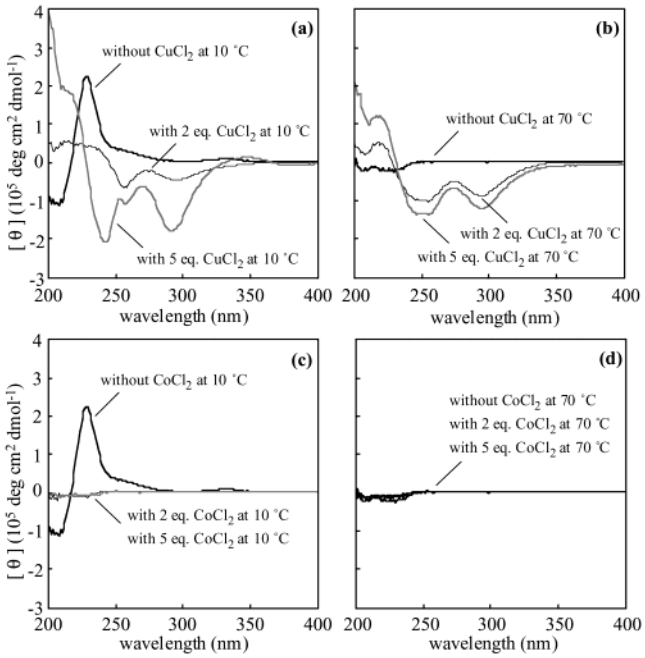

Figure 6. Effect of concentration of metal chlorides on CD spectra with 0.3 mM **1** in cyclohexane−ethanol (100:1) solutions.

70 °C (Figure 6d). These results indicate that the complexation with $CuCl_2$ promotes production of supramolecular aggregates that are different from the original aggregates but the complexation with $CoCl_2$ is not comfortable to maintain highly ordered structures and rather perturbs to destroy them. A similar effect to that of $CoCl_2$ was observed for $ZnCl_2$.

Effect of Metal Chlorides on Aggregation Morphology. It was confirmed that the aggregation morphology was also influenced by metal chlorides. TEM observation showed that addition of 1 equimolar of $CoCl_2$ destroyed the fibrillar networks, causing them to be just fragmented (Figure 7a). On the other hand, in the case of $CuCl_2$, they were fragmented but developed tapelike aggregates (Figure 7b). This different action is related to the fact that the complexation with $CuCl_2$ induces chirally ordered structures different from the original ones but $CoCl_2$ only destroys them as indicated by the CD spectra.

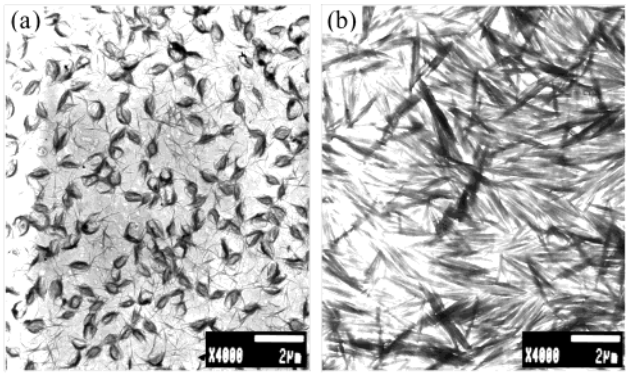

Figure 7. Transmission electron micrographs of aggregates of **1** in cyclohexane−ethanol (100:1) solutions: (a) with $CoCl_2$; (b) with $CuCl_2$. Initial concentrations: [**1**] = 0.3 mM; [metal chlorides] = 0.3 mM.

In conclusion, not only can **1** form nanofibrous aggregates based on highly ordered structures in organic systems, but also the aggregation morphology and the chirality can be controlled by complexation with metal chlorides at the headgroups. The different actions recognizing the kind of metal chlorides are probably derived from those of coordination structures between Cu and Co. Here, Cu is a typical metal forming a square planar coordination state but Co and Zn form an octahedral coordination state. It is in no doubt that the octahedral structures do not keep their orientation among lipids although their exact structures are not yet specified. On the other hand, a square planar structure by Cu is surely comfortable to keep its molecular orientation while the original structure is not maintained. Here, we have established a method to control the chirality of organogels by metals, and thus it promises to expand possible applications.

Acknowledgment. This research was supported in part by a Grant-in-Aid for Scientific Research from the Ministry of Education, Science, Sports, and Culture of Japan.

LA025535F