

Studies of Helical Aggregates of Molecules. I. Enantiomorphism in the Helical Aggregates of Optically Active 12-Hydroxystearic Acid and Its Lithium Salt

Taro TACHIBANA and Hideko KAMBARA

Department of Chemistry, Ochanomizu University, Bunkyo-ku, Tokyo

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It was found, by electron-microscope observation, that D(or L)-12-hydroxystearic acid and its Li soap crystallize from a solution in various solvents in the form of a twisted fiber. The twist was left-handed for the D-acid and right-handed for the L-acid, while the Li soap gave results just opposite to those of the corresponding acid. The DL-acid separated out not fibers, but plate-like crystals, and its Li soap produced non-twisted fibers. Thus, an enantiomorphic relationship for the sense of twist between the D-enantiomer and the L-enantiomer was found both for the acid and for the Li soap.

Spirals or helices are often seen in various biological systems (*e.g.*, chromosome, bines, and snail shells). Curiously, some biological helices have a definite sense; they have either a right-handed or left-handed sense, depending upon the kind of biological system. Though one may imagine that this fact is related to the asymmetry of biological molecules, no evidence has yet been presented for such a speculation. Interest in this problem has lead us to notice the sense of twist in the helical aggregates of molecules.^{1,2)}

It has been known by electron-microscope study, that some crystalline aggregates of soap from lubricating grease consist of fibers in the form of twisted ribbon or helical rope. On examining the electron micrographs of the fibers hitherto published, we found that the fibers from hydrated calcium tallo-water grease exhibit both right-handed and left-handed twists, while the fibers from lithium 12-hydroxystearate grease all twist by the same hand. This fact had also been pointed out by Hotten and Birdsall,³⁾ but they did not report the sense of twist of the lithium soap fibers. We found in the literature that some micrographs^{4a-c)} exhibit a right-handed twist and others,^{3,5a,b)} a left-handed twist. There is, then, confusion in the literature, but un-

doubtedly either sense of the twist could occur in a specimen. Since 12-hydroxystearic acid is an optically-active compound and since the naturally-occurring enantiomer is the D-form, the molecular asymmetry may be reflected in the sense of twist. In the present investigation, we have undertaken to determine the sense of twist in the specimens and to confirm whether or not there exists an enantiomorphic relationship between the helical aggregates from each enantiomer.

Experimental

Materials. D-12-Hydroxystearic acid was purified by the urea-adduct method from a commercial product containing as impurities about 15% stearic and palmitic acids. This procedure consists of dissolving the methyl ester into urea-saturated methanol with warming, letting the solution stand at room temperature, and then precipitating the adduct. Since the adduct initially precipitated was rich in methyl stearate and palmitate, they were removed by filtration. The methyl hydroxystearate-rich adduct was obtained by adding further urea to the filtrate. The adduct was decomposed with water, thus isolating the methyl hydroxystearate. The methyl hydroxystearate was repeatedly purified by the urea-adduct method. The final product was over 99% pure. The acid was obtained by saponifying the methyl ester with alcoholic KOH and by then acidifying it with dilute H₂SO₄; mp 79.3–79.8°C, $[\alpha]_D^{25}$ –0.3 ± 0.1° (pyridine); lit⁶⁾ mp 80.5–81°C, $[\alpha]_D^{25}$ –0.41° (pyridine).

DL-12-Hydroxystearic acid⁷⁾ was obtained by the reduction of 12-ketostearic acid derived from D-12-hydroxystearic acid.

6) K. Serck-Hanssen, *Chem. Ind. (London)*, **1958**, 1554.

7) I. Nakajima and S. Toyama, *Nippon Kagaku Zasshi (J. Chem. Soc. Japan, Pure Chem. Sect.)*, **81**, 1598 (1960).

1) T. Tachibana and H. Kambara, *J. Am. Chem. Soc.*, **87**, 3015 (1965).

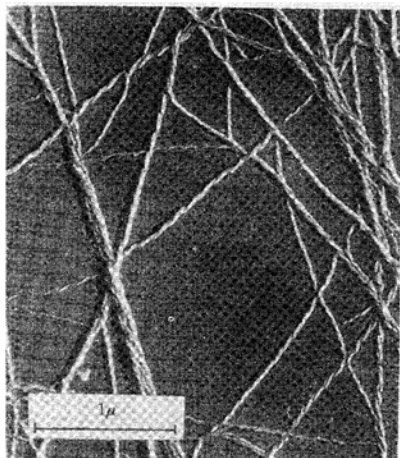
2) T. Tachibana and H. Kambara, *J. Colloid Interfac. Sci.*, **28**, 173 (1968).

3) B. W. Hotten and B. H. Birdsall, *J. Colloid Sci.*, **7**, 284 (1952).

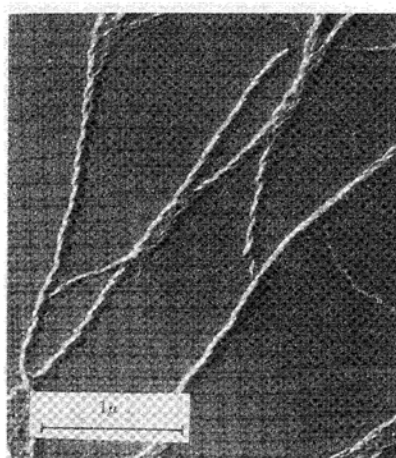
4) a) A. Bondi, A. M. Cravath, R. J. Moore and W. H. Peterson, *NLGI Spokesman*, **13**(12), 12 (1950); b) B. W. Hotten, *ibid.*, **19**(1), 14 (1955); c) G. V. Vinogradov and V. V. Sinitsyn, *J. Inst. Petrol.*, **47**, 357 (1961).

5) a) M. J. Vold, V. A. Elersich, R. F. Baker and R. D. Vold, *NLGI Spokesman*, **18**(5), 8 (1954); b) A. L. McClellan, *J. Chem. Phys.*, **32**, 1271 (1960).

(A) 12-Hydroxystearic Acid

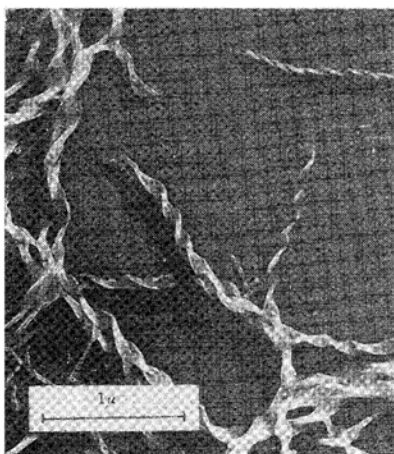


(a) D-form, left-handed twist

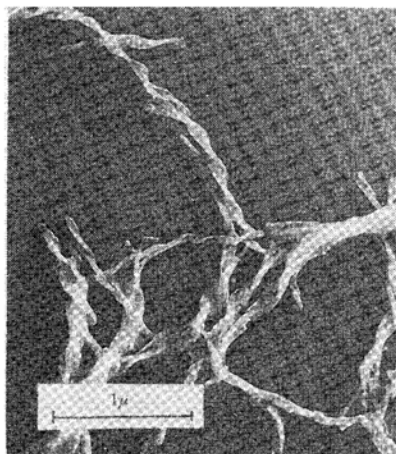


(b) L-form, right-handed twist

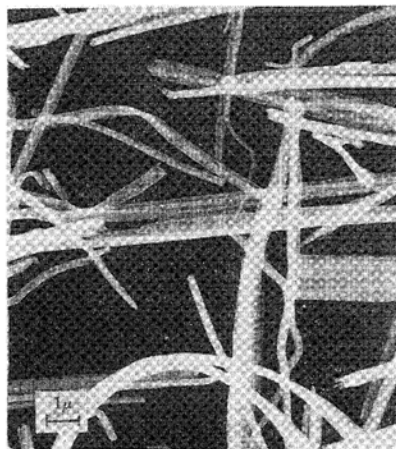
(B) Lithium 12-Hydroxystearate



(a) D-form, right-handed twist



(b) L-form, left-handed twist



(c) DL-form, no twist

Fig. 1. The effect of optical isomerism on the sense of the twist in fibrous aggregates of 12-hydroxystearic acid (A) and lithium 12-hydroxystearate (B).

L-12-Hydroxystearic acid was prepared by the Walden inversion of the D-acid. The methyl ester of the D-acid was tosylated with *p*-toluenesulfonyl chloride in pyridine at a low temperature. The tosylate was then converted to the acetate by refluxing it with sodium acetate in acetic acid containing a small amount of acetic anhydride. The Walden inversion then occurred. L-12-Hydroxystearic acid was obtained by saponifying the acetate with KOH, followed by acidification with dilute H_2SO_4 and purification by recrystallization from methanol and then from acetone; mp 75.6–76.0°C, $[\alpha]_D^{25} +0.40 \pm 0.05^\circ$ (pyridine), lit.⁽⁶⁾ mp 80.4–80.6°C and 78°C (dimorphous), $[\alpha]_D^{25} +0.3 \pm 0.1^\circ$ (pyridine). The infrared spectra of this L-acid was identical with that of the D-acid.

The analysis of the hydroxy acids was done by gas chromatography after they had been converted into the methyl ester of the acetoxy acid.

The Li soap was prepared by neutralizing the acid with LiOH in an ethanol solution, then recrystallizing the acid several times from ethanol, and finally drying it over P_2O_5 *in vacuo*.

The Formation of the Aggregate. The method consists of dissolving the soap into a hot solvent and then allowing it to stand at room temperature. The solution then solidifies into grease or gel, or separates out a gel-like precipitate, depending on the concentration and on the nature of the solvent used. The solvents were a highly refined mineral oil, Nujol, and ethanol. In a similar manner, 12-D(or L)-hydroxystearic acid itself was also found to form a helical aggregate from Nujol, benzene, carbon tetrachloride, chloroform, acetone, and ethanol.

Electron-microscope Observation. In order to make a specimen for the electron microscope, a small portion of the gel was placed on a copper grid covered with a thin film of carbon-collodion; the solvent was then evaporated *in vacuo*. When an involatile solvent, such as mineral oil or Nujol, was used, a small portion of the gel was dispersed in solvent naphtha, in which the dispersed phase is insoluble. Then a small drop of the dispersion was placed on the grid and dried. After that, the specimens were shadowed with Pt-Pd metal. The observations were done with a JEM-T6 electron microscope (direct magnification, X 30000).

In order to determine the helical sense from an electron micrograph, some care must be taken. The helical sense appearing in the electron micrograph depends on whether the specimen shadowed previously with metal was exposed to the electron beam directly or through a support film, since only the metal-shadowed side of a helix gives the electron-microscope image clearly. Accordingly, a right-handed helix appears as either a right-handed image or a left-handed image, depending on the mode of the exposure of the specimen to the electron beam. Consideration should be also given to the process of making a copy of the photograph. These factors seem to us to have escaped earlier workers' notice. The photographs in this paper are printed to show the same sense as that of the twist in the specimen.

Results and Discussion

Some typical micrographs are shown in Fig. 1, in which the twisted fibers are clearly visible. It can also be seen that the structure is actually made

up of two or more individual fibers twisted about one another. The widths of the fibers varied over a considerable range.

The hydroxy acid was found to form a twisted fibril 0.01–0.1 μ in width, regardless of the kind of solvent used. Sometimes the fibers from benzene and from carbon tetrachloride were several tens of microns long.

In the case of the lithium soap, twisted fibrils about 0.03 μ in width were formed from ethanol, while helical ribbons 0.03–0.3 μ in width were formed from mineral oil and from Nujol. The thickness of the ribbon was so thin that, when a ribbon was laid upon another ribbon, the lower ribbon was visible through the upper. This was observed clearly when the specimen was lightly shadowed with metal. The thickness of the ribbons appeared to be about 0.01 μ ; however, the value could not be estimated accurately from our micrographs. When very thin ribbons are separated out from a nonpolar solvent, they should develop a nonpolar surface. Therefore, it may be plausible to assume that the soap molecules are oriented along the direction of the thickness of the ribbon, exposing the methyl groups. In this case, the thickness corresponds to approximately the length of four molecules, and the width, to from several tens to several hundreds of molecules. In general, these ribbon-like fibers have a tendency to increase in width rather than in thickness by molecular association. The twisting is more clearly seen in a thinner fiber or a narrower ribbon.

One of the parameters of a helix is the sense of twist, to which we have particularly directed our interest in this study. The sense of twist is always definite for each enantiomer, regardless of the experimental conditions. The results are presented in Table 1. The sense of twist is left-handed for the D-acid and right-handed for the L-acid. On the other hand, the lithium soap gave results just opposite to the corresponding acid. The DL-acid (the racemic form) separated out not fibers, but plate-like crystals, and its lithium soap produced non-twisted fibers. Thus, an enantiomorphous relationship for the sense of twist between the D-enantiomer and the L-enantiomer was established for the acid as well as for the lithium soap.

It is a problem for further study what mechanism serves to twist the fibers. As is well known, twisted

TABLE 1. THE SENSE OF TWIST IN THE FIBERS FROM BOTH ENANTIOMERS OF 12-HYDROXYSTEARIC ACID AND THEIR LITHIUM SOAPS

Enantiomer	The sense* of twist	
	Acid	Li soap
D	l	r
L	r	l

* r: right-handed twist, l: left-handed twist.

fibers are also formed from hydrated calcium talowate grease. They consist of fibers with a right-handed twist and fibers with a left-handed twist in the same proportion. This indicates that, when optically non-active molecules aggregate to form twisted fibers, they exhibit both the sense of twist with an equal probability.

Several investigators^{8a, b)} have observed that the grease soap fibers have distinctly less twist without free end when prepared by a gentle deoiling manipulation, and that twisted fibers with a free end are often exhibited in cases involving the deformation of the gel structure by mechanical force during deoiling. Thus, it has been concluded that the twisted form is probably the result of mechanical breakdown. These investigations have been made with interest directed to the gel structure in lubricating grease. However, it is noticeable, from the molecular point of view, that the fibers exhibit the twisting when the fibers with a free end are produced. This fact shows also that the twisting occurs not only when the fibers grow, but also after they were formed. This indicates that the

the isolated fiber has in itself a tendency to twist spontaneously. The enantiomorphism found for the sense of twist may also be considered as evidence that the twisting involves a molecular mechanism.

Here it seems appropriate to cite a paper by Weitkamp,⁹⁾ who observed that the dextro-rotatory anteiso acids $(\text{CH}_3\text{CH}_2\text{CH}(\text{CH}_3)(\text{CH}_2)_{2n}\text{COOH})$ from degrease crystallize from solution in mineral oil in a tubular form which is generated by the counterclockwise spiral growth of a very thin and quite narrow ribbon. If the levo-rotatory acid is employed, the clockwise spiral growth of the ribbon will be observed.

As is well known, single crystals of optically-active antipodes exhibit the phenomenon of enantiomorphism. It is interesting to note that the enantiomorphism has been found to occur in the crystalline aggregate when the optical isomer aggregates to possess helical symmetry.

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8) a) M. J. Vold, V. A. Eilersich, R. F. Baker and R. D. Vold, NLGI Spokesman, **18**(5), 8 (1954); b) A. L. McClellan and J. Cortes, *ibid.*, **20**(6), 12 (1956); c) J. M. Wilson, Jr., *ibid.*, **27**(12), 372 (1964); d) F. W. Anderson, R. C. Nelson and F. F. Farley, *ibid.*, **31**(7), 252 (1967).

9) A. W. Weitkamp, *J. Am. Chem. Soc.*, **67**, 447 (1945).