BL-10C/2023G547

Nucleotide-based fibers comprising ion complex between biomass DNA and cationic lipid

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1 Introduction

Developing technology to produce high-performance fibers from bio-derived waste is an important issue from the perspective of SDGs. Salmon, *Oncorhynchus keta,* is a common fishery resource estimated to have a worldwide supply exceeding several million tons annually. A large number of salmon fillets and eggs, as well as canned salmon, are produced in Japan. However, most salmon milts are discarded at an approximate amount of several thousand tons annually because they degrade easily and are not suitable for food. Alternatively, salmon milt contains a large DNA amount. Moreover, nucleic acids are generated on the order of approximately 3.5 million tons per year as a yeast fermentation byproduct. DNA is highly water-soluble and biochemically unstable. These properties have been making it difficult to utilize them as industrial and/or functional materials. For overcoming this weak point, DNA as a molecular material has recently been studied regarding complexation with cationic lipids. Even though the synthetic lipids are often toxic to the living cells, the conversion of these discarded DNAs into useful materials would be beneficial to utilize these unique natural resources. If it is possible to convert salmon milt into regenerated fibers and put them into practical use, effective resource utilization will be realized, contributing to bio-based fiber production.

In this study, we produced biomass DNA-based regenerated fibers comprising ion complexes between cationic lipids and DNA molecules extracted from salmon milt waste (Fig.1). The DNA–lipid complex was soluble in ethanol, which was used as a dope solution using the dry spinning method. The mechanical properties were maintained regardless of the drawing extension degree. The conformational structure of the DNA–lipid fibers was influenced by humidity, and the DNA molecules were randomly oriented irrespective of the drawing extension. The fibers obtained in this study can be applied not only to clothing and filters but also to reversible switching materials that change their structure depending on humidity. Furthermore, DNA–lipid complex displays a large surface area and intercalation ability, contributing to chelating harmful cationic or aromatic compounds released into the ocean.

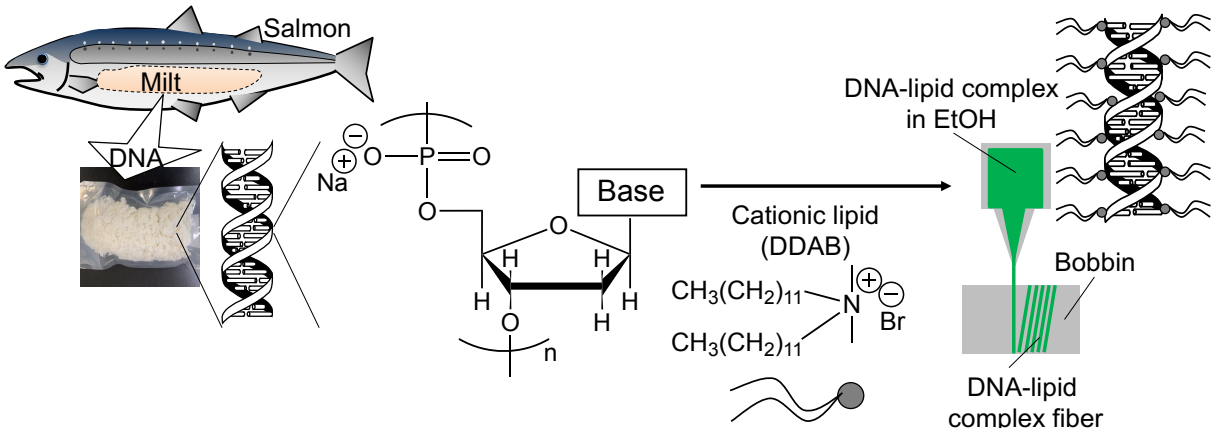

Fig.1. Dry spinning of ion complexes between cationic lipids and DNA molecules that were extracted from biomass salmon milts. After the ion complex formation with a quaternary cationic surfactant (DDAB), the electrostatic repulsions between the phosphate anions decreased, which was key to successful spinning.

2 Experiment

The wide-angle X-ray scattering (WAXS) measurements of the DNA fibers were performed with a 0.1 nm wavelength at the beamline, BL-10C, in Photon Factory, Tsukuba, Japan. The sample-detector distance was set to 232 mm. The DNA–lipid fiber samples were stored under RHs of 0% or 97% before measurement. The obtained scattering data were radially integrated to convert two-dimensional profiles into one-dimensional ones using the software Fit2D. In addition, the *d*-spacing value was calculated based on the following equation (1).

$$d = \frac{2\pi}{q} \quad (1)$$

where d is the interplanar spacing and q is the absolute scattering vector value obtained from the one-dimensional profile.

3 Results and Discussion

WAXS measurements can monitor the coherent interference of scattered X-rays passing through partially ordered materials. This enables examining the repetitive distances for molecular packing and lamella structures. WAXS measurements were conducted to further investigate the molecular packing and conformational structure of the DNA–lipid fibers. Two-dimensional WAXS profiles for the DNA–lipid fibers under drawing extension were examined. The WAXS profiles had concentric scattering rings, namely Debye-Scherrer rings, indicating that the DNA–lipid fibers were randomly oriented. The two-dimensional WAXS data were converted into one-dimensional WAXS data by radial integration. Based on the one-dimensional WAXS profiles, the molecular packing and conformational structures of the DNA–lipid fibers were maintained after drawing extension. The WAXS peak corresponding to the *d*-spacing value of 3.2 or 3.3 nm was detected. This peak suggests the repeat distance of the lamella structure comprising single-stranded DNA with a 1.0 nm thickness and lipid bilayers with a 2.3 nm thickness, according to the literature for the DNA–cationic lipid films. The WAXS profile of DDAB molecules was used as a reference. Generally, polymeric crystals exhibit broader interference fringes, whereas crystals comprising low molecular weight compounds exhibit sharper peaks during WAXS measurements. The sharper peaks in the one-dimensional profile should be derived from DDAB molecules. The WAXS profiles exhibited broader interference fringes, indicating that DDAB molecules formed ion complexes with DNA molecules.

A broad peak was also detected at the *q*-value of 14 nm^{-1}, corresponding to the *d*-spacing value of 0.43 nm. The peak broadness suggests the scattering of the *d*-spacing value. From the literature, it is known that discotic liquid crystals exhibit a broad WAXS peak corresponding to the *d*-spacing of 0.43 to 0.47 nm due to the presence of long alkyl chains. Therefore, the broad peak detected here most likely originated from the packing distance of long alkyl chains of the DDAB molecules.

Next, we investigated the humidity-dependent structural change of the DNA–lipid fibers. We selected the DNA–lipid fibers obtained by drawing extension of 2 as an example among the four samples with the different drawing extension ratios. We obtained the WAXS data when the DNA–lipid fibers were immersed in water and subsequently dried. The peak with the *d*-spacing of 4.3 nm was detected under wet conditions. The *d*-spacing value suggests the repeat distance of the lamella structure comprising double-stranded DNA molecules with a 2.0 nm thickness and lipid bilayers with a 2.3 nm thickness based on the literature for the DNA–cationic lipid films. Subsequently, the DNA–lipid fibers were dried, and the peak shifted back to the *d*-spacing of 3.3 nm.

A previous study reported that stretching DNA–lipid films in water improves molecular orientation. By contrast, the molecular orientation of DNA–lipid fibers did not change significantly upon stretching in this study. A reasonable explanation is that the molecular weight of the DNA used in this study was smaller, ranging from 100 to 700 bp, based on the agarose gel electrophoresis. This is in contrast to the molecular weight of 2000 bp in the previous study. The lower molecular weight led the lack of molecular entanglement, and the molecular chains might not be stretched and were randomly oriented in the DNA–lipid fibers. Increasing temperature will induce the dissociation of hydrogen bonds between DNA backbones, and double-stranded DNA molecules transform into single-stranded states. Additionally, increasing humidity is also known to cause structural transition of DNA from single-stranded to double-stranded states at 25 °C according to a previous report. In this study, we observed the structural transition of DNA molecules at 25 °C. Thus, the structural transition was induced not by temperature but by humidity.

Acknowledgement

This work was financially supported by JSPS Grant-in-Aid for Scientific Research (C) Grant No. 21K12305.

References
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