STRUCTURAL ANALYSIS OF HUMAN HAIR CUTICLE USING MICROBEAM X-RAY DIFFRACTION: RELATIONSHIP WITH EFFECTS OF HAIR DYEING

Hair treatments, such as permanent waving, hair bleaching and hair conditioning, are performed in wet conditions. Thus, in cosmetic research, it is important to understand how water-soluble products can penetrate into a hair fiber. The cuticle is the outermost layer of a hair fiber and is made up of a stack of several sheet-like cells (Fig. 1). This structure is thought to have a large effect on the permeation of substances into hair. The surface of the cuticle cell is covered by a thin layer of lipids called β -layer. Between adjacent cells, these lipids are separated by the δ -laver of proteinous components. This triplelayered structure between adjacent cells is called a cell membrane complex (CMC). CMC is the only substructure that continuously fills the intercellular spaces of a hair fiber. Therefore, it is considered to be an important pathway for the penetration of substances.

Recently, small angle X-ray scattering (SAXS) analysis using a microbeam has been reported to be useful for characterizing CMC structure [1]. We have characterized human hair samples using SAXS analysis and have detected changes in CMC structure that correlate with the penetration of molecules [2].

Using a 5- μ m ϕ high flux beam (λ =0.083 nm), SAXS experiments were carried out at beamline **BL40XU** under an atmosphere of 30 °C and 50% relative humidity. Each thickness of the β -layer and δ -layer was estimated from X-ray scattering patterns using an electron density model [3]. For SAXS experiments, hair strands were obtained from Japanese women who had not used any chemical treatments such as permanent waving or hair bleaching. The strands were subjected to extraction with four different solvents, namely, methanol, acetone, hexane, and a chloroform/methanol mixture (2:1 v/v), at 37 °C for 6 hours. Because was reported that solvent extraction accelerated the dyeing rate of wool fiber [4], we planned to detect the influence of these extraction treatments on CMC structure.

The estimated thickness of the CMC structure of the hair samples is shown in Fig. 2. The thickness of the β -layer was decreased by extraction with acetone and hexane, while it was not changed by extraction with methanol or chloroform/methanol. In addition, the thickness of the δ -layer was decreased by extraction using methanol, acetone, and chloroform/methanol, and did not significantly change with hexane. Interestingly, these hair samples differed in the dyeing extent of Acid Orange 7 as an index of substance penetration.

Figure 3 shows the relationship between the CMC structure and the dyeing extent. There is a correlation between the thickness of the δ -layer and the dyeing extent, the larger decrease in δ -layer thickness resulting in a greater elevation of the dyeing extent. On the other hand, the thickness of the β -layer did not show a correlation with the dyeing extent. These results suggest that the δ -layer acts as a major pathway of acidic dye penetration.

SAXS analysis using an X-ray microbeam is a powerful tool for characterizing the CMC structure in the area of cosmetic research. The main advantage



Fig. 1. Transmission electron micrographs of human hair and schematic diagram of CMC structure.

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Industrial Applications





of this method over transmission electron microscopy is the availability of hair analysis in natural conditions, as the hair samples do not require fixing, pre-staining or slicing. Therefore, we were able to detect structural changes in the CMC by solvent extraction. Fixing and pre-staining treatments would induce serious chemical changes of hair components that are comparable to the solvent extraction.

Our findings showed a correlation between dyeing extent and δ -layer thickness, both of which were

changed by extraction with solvents. It has been speculated that hydrophilic molecules penetrate hair through the δ -layer, on the basis of histochemical observations of the CMC. Since the dye used in our study (Orange 7) was water-soluble, the relationship seen between the dyeing extent and the δ -layer thickness is in agreement with this speculation. Thus, we were able to detect changes in the CMC structure that correlated with the penetration of molecules using a microbeam SAXS method.



Fig. 3. Relationships between CMC structure and extent of dyeing. (a) Relationship between thickness of β -layer and dyeing extent. (b) Relationship between thickness of δ -layer and dying extent. ΔE ; index of dyeing extent measured using a chromometer, R; correlation coefficient, p; level of significance.

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