

Effect of transdermal absorption enhancers on the lipid lamellar structures in stratum corneum

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Introduction

Systemic delivery through the skin is difficult for most drugs because the stratum corneum has a barrier function. When effective transdermal delivery of diclofenac was sought, *L*-menthol was considered to be a promising facilitator of permeation. The molecular arrangement of intercellular lipids in the stratum corneum has been investigated in the hairless mouse using synchrotron X-ray diffraction [1]. Synchrotron X-ray diffraction profiles of the rodent stratum corneum were therefore considered to be valuable in understanding changes in molecular arrangements of intercellular lipids caused by absorption enhancers. This technique may be used to clarify the molecular mechanisms whereby enhancers promote drug permeation.

Materials and Methods

The stratum corneum was separated from skin of the abdominal regions of hairless rats (strain HWY/Slc, 12 weeks old, Sankyo Labo Service, Japan) by digestion with 0.1% (w/w) trypsin in phosphate-buffered saline (pH 7.4) at 37 °C. The separated sheet of stratum corneum was rinsed in distilled water and dried in vacuo. Weighed stratum corneum (ca. 5 mg) was soaked in a solution containing absorption enhancer for a fixed time. The treated stratum corneum was then dried under a nitrogen gas stream until the expected weight (125% of weight prior to treatment) was achieved. X-ray diffraction was performed using a monochromatic synchrotron beam at Station BL15A in the Photon Factory (Ibaraki, Japan). The wavelength (λ) of the X-ray beam was 0.1506 nm. The sample-to-detector distance was about 150 mm. The reciprocal spacing (S) was calibrated from the spacing of silver behenate, 58.38 nm at 25 °C. The value of S is given by: $S=n/d=(2/\lambda) \sin (\theta/2)$ where 2θ is the scattering angle and d is a repeat distance. A capillary tube with a diameter of 1 mm containing prepared stratum corneum was sealed by a gas burner and placed in a sample holder of the X-ray diffractometer. The sample was kept at room temperature throughout the experiment. The X-ray diffraction profiles were recorded with an imaging plate (Type BAS-III, Fuji Photo Film Co. Ltd., Tokyo, Japan). Diffraction intensity was read on a BAS2500 system (Fuji Photo Film Co. Ltd., Tokyo, Japan).

Results and Discussion

The profile of the hairless rat stratum corneum using synchrotron X-ray diffraction in the wide-angle region is shown (Fig. 1). Two diffractions ($S=2.40 \text{ nm}^{-1}$ and $S=2.67$

nm^{-1} , corresponding to spacing of 0.417 nm and 0.374 nm, respectively) were observed on the broad hump peak derived from soft keratin. These diffractions indicated that the hairless rat stratum corneum lipids were organized in both hexagonal and orthorhombic hydrocarbon chain packing as are those of the hairless mouse and humans. The apparent abundance ratio of hexagonal/orthorhombic lipid hydrocarbon chain packing ($R_{H/O}$) was defined as follows:

$R_{H/O}=(\text{Peak area at } S=2.40 \text{ nm}^{-1}/\text{Peak area at } S=2.67 \text{ nm}^{-1})$. When ethanol was applied to the stratum corneum the intensities of diffraction declined slightly. In X-ray diffraction of stratum corneum, declines in intensity indicate decreases in the amounts of ordered lipids in the stratum corneum. $R_{H/O}$ value was not significantly affected by ethanol concentration in the range 0–40% (w/w). When *l*-menthol was used at 2% (w/w), a significant decrease of $R_{H/O}$ value was observed (Fig. 2). The observed decrease in $R_{H/O}$ value suggested that treatment with 2% (w/w) *l*-menthol preferentially altered the lipid hexagonal hydrocarbon chain packing. The change in the $R_{H/O}$ value might affect the barrier ability of stratum corneum to drug permeation because of the possibility of decreasing dense barrier formed by lipid hydrocarbon chain packing [2].

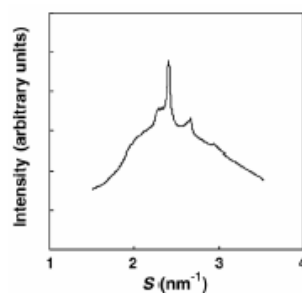


Fig.1 Wide-angle X-ray diffraction profile of hairless rat stratum corneum.

References

- [1] N. Ohta et al., Chem. Phys. Lipids, **123**, 1-8 (2003).
 [2]. Y. Obata et al., J. Contr. Rel., **115**, 275-279 (2006).

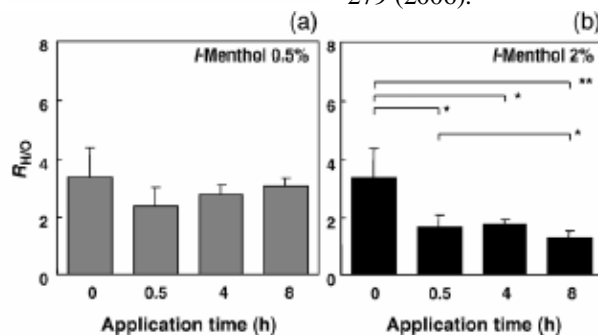


Fig.2 Effect of concentration of *l*-menthol and application times on lipid hydrocarbon chain packing of stratum corneum

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