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The Change and Recovery of Human Skin Barrier Functions after Ultraviolet Light Irradiation

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The changing patterns of *in vivo* human skin barrier functions after irradiation with a single middle-wavelength ultraviolet light (UV-B) were determined with respect to trans-epidermal water loss (TWL) and skin surface lipid values.

Quantitative results were obtained as follows. 1) A significant increase of the TWL value was found 1 week after UV-B exposure at the site irradiated with twice the minimal erythemal dose (MED). 2) Cholesterol value in skin surface lipid was raised significantly 2 or 3 weeks after UV-B exposure at the 2 MED site. On the other hand, total lipid and squalene values showed no significant differences. 3) It was noteworthy that the TWL value recovered after 3 weeks to the initial control level at all UV-B energy doses. Similar tendencies were observed in the cases of total lipid, squalene and cholesterol values. 4) The increase of energy dose was parallel to the increase of the TWL value, *i.e.* lowering of the barrier efficacy of the *stratum corneum*.

Keywords——human skin barrier functions; middle-wavelength ultraviolet light; TWL; skin surface lipids; squalene; cholesterol; change and recovery pattern of TWL and lipids; energy dose-barrier efficacy

In the literatures,[2] it is assumed that human skin barrier functions are impaired by ultraviolet light (UV) exposure.

Alterations of these functions may have important implications with respect to the study of the optimum use of external protective preparations against UV irradiation or that of changes in the physical state of the skin surface.[3] In this regard, especially for sunburn treatment products, it is essential to understand skin barrier efficacy changes after UV irradiation.

However, no quantitative data on the effect of UV irradiation on the human skin barrier functions has been found in recent years. Therefore, the present study was carried out to obtain basic knowledge of human skin surface barrier changes after a single middle-wavelength UV (UV–B) exposure.

Thus, transepidermal water loss (TWL) and skin surface lipid values, both of which were shown to be important parameters in skin surface barrier properties,[4] were determined with time after UV–B irradiation.

The purpose of this paper is to report typical changing patterns of TWL and skin surface lipid values. In addition, the relationships between irradiated energy doses and barrier efficacy changes are discussed.

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Experimental

Subjects——Ten healthy male adults aged between 21 and 54 years were the subjects of this study.

UV-B Irradiation——Each volunteer recieved UV-B irradiation on the back in a single exposure. The UV light was emitted by four tubes (Toshiba FL-20SE) which emitted 290—400 nm, with the maximum energy at 305±5 nm. It is generally recognized that sunburn is produced mainly by the UV-B (280—315 nm) spectrum.[2a,c]

Minimal erythemal dose (MED) represents that amount of energy producing a barely perceptible sunburn or erythema of the skin.[2a,c] On the basis of each individual MED irradiation time, three energy level irradiations (1/2, 1 and 2 MEDs) were performed at a distance of 20 cm from the lamps. Each irradiated area was 9.0 cm^2. The mean irradiation time for 1/2 MED was 1 min, that for 1 MED was 2 min and that for 2 MED was 4 min.

The energy delivered at the skin surface was determined by a 0.006 M potassium ferrioxalate chemical actinometer[5] to be 2.24×10^4 $erg/cm^2/sec$ at 300 nm. From this, the mean energy for 2 MED was 5.4×10^6 erg/cm^2, that for 1 MED was 2.7×10^6 erg/cm^2 and that for 1/2 MED was 1.4×10^6 erg/cm^2.

TWL Measurements——TWL measurements were performed on each irradiated site at 0 hr, 24 hr, 1 week, 2 weeks and 3 weeks after a single UV irradiation.

TWL was measured by the method of electrohygrometry previously reported[4] and TWL values were expressed as $mg/cm^2/hr$. The skin surface temperature was measured with a thermister thermometer. Measurements were carried out at 21—23° and 60—70% relative humidity.

Lipid Analysis——Casual lipids were extracted with acetone by the cup method (7.0 cm^2 in area) from the only 2 MED irradiated site. Replacement lipids were obtained 2 hr afterward. Collections were made at 0, 24 hr, 1, 2 and 3 weeks intervals.

The collection procedure was the same as that previously reported.[4] Total lipids were measured gravimetrically. Free and total cholesterol and squalene were analyzed by gas chromatography. The quantitative gas chromatographic technique described earlier[4] was applied to the lipid samples obtained.

Results

Changes in TWL Value after UV-B Irradiation

The changes in TWL value are shown in Table I. A statistical difference was found only in the significant increase of TWL values after 1 week at the 2 MED irradiated site ($p<0.05$).

To understand the relation between irradiated energy doses and TWL values, the percent change of the TWL value at each irradiated energy dose is illustrated in Fig. 1.

TABLE I. Changes in TWL Value after UV-B Irradiation

Time	TWL values ($mg/cm^2/hr$)		
	1/2 MED	1 MED	2 MED
0 hr	0.215±0.030[a] (31.3±0.7)[b]	0.210±0.042 (31.4±0.6)	0.225±0.039 (31.3±0.5)
24 hr	0.221±0.025 (31.5±0.6)	0.214±0.026 (31.5±0.7)	0.223±0.035 (31.5±0.6)
1 week	0.221±0.026 (31.0±0.2)	0.244±0.052 (31.1±1.0)	0.282±0.059[c] (31.2±1.0)
2 weeks	0.223±0.040 (31.4±0.5)	0.228±0.035 (31.5±0.5)	0.258±0.058 (31.6±0.7)
3 weeks	0.210±0.039 (30.7±1.0)	0.213±0.042 (30.7±0.8)	0.217±0.034 (30.8±0.8)

Mean energy was 1.4×10^6 erg/cm^2 for 1/2 MED, 2.7×10^6 erg/cm^2 for 1 MED and 5.4×10^6 erg/cm^2 for 2 MED.
a) Mean±S.D. of TWL value of 10 volunteers.
b) Mean±S.D. of skin temperature of 10 volunteers.
c) $p<0.05$; significantly different from unirradiated control

5) C.G. Hatchard and C.A. Parker, *Proc. Roy. Soc.*, **A235**, 518 (1956).

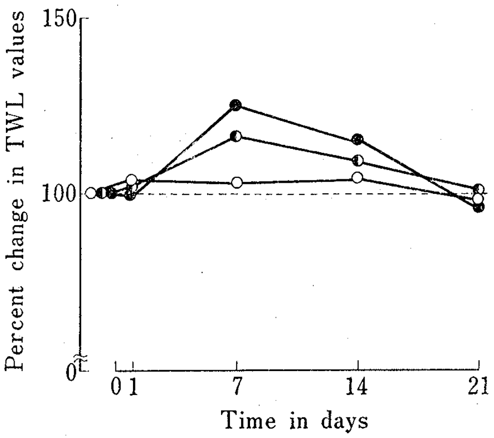

Fig. 1. Time Course of Changes in TWL Value after UV–B Irradiation

The mean percent change was calculated on the basis of the mean value at various days after irradiation.
—○—: 1/2 MED; 1.4×10^6 erg/cm^2.
—◐—: 1 MED; 2.7×10^6 erg/cm^2.
—●—: 2 MED; 5.4×10^6 erg/cm^2.
------: unirradiated control level.

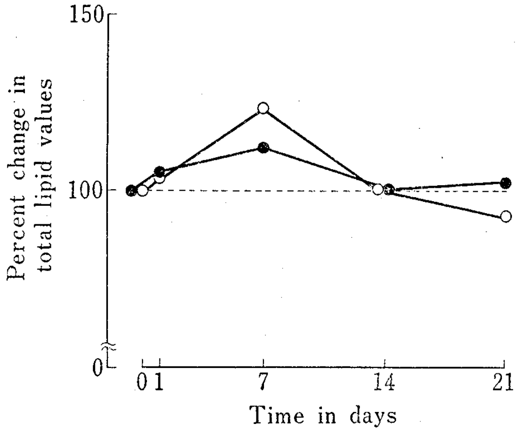

Fig. 2. Time Course of Changes in Total Lipid Value after UV–B Irradiation

The mean percent change was calculated on the basis of the mean value at various days after irradiation.
—○—: casual lipids.
—●—: replacement lipids for 2 hr.
------: unirradiated control level.
UV-B dose: 5.4×10^6 erg/cm^2 (2 MED).

Table II. Changes in Lipid Value after UV–B Irradiation

Time	Total lipids (μg/cm²)		Squalene (μg/cm²)		Free cholesterol (μg/cm²)		Total cholesterol (μg/cm²)	
	C.L.[a]	R.L.[b]	C.L.	R.L.	C.L.	R.L.	C.L.	R.L.
0 hr	76.50± 31.12[c]	47.35± 21.16	1.17± 0.72	0.86± 0.92	0.61± 0.38	0.43± 0.19	1.18± 0.95	0.55± 0.15
24 hr	78.46± 38.84	49.93± 22.20	1.30± 0.81	0.49± 0.34	0.75± 0.33	0.53± 0.13	1.57± 0.36	0.62± 0.19
1 week	94.45± 35.30	53.30± 17.65	1.57± 0.55	0.72± 0.23	0.82± 0.66	0.56± 0.46	1.84± 1.75	0.88± 0.37
2 weeks	76.50± 36.57	47.45± 11.82	1.14± 0.68	0.55± 0.39	1.78± 1.01[d]	0.88± 0.82	2.55± 2.02[e]	1.37± 0.97[f]
3 weeks	70.38± 17.65	48.24± 5.80	1.26± 0.78	0.88± 0.81	0.79± 0.35	0.61± 0.17	1.89± 1.33	1.16± 0.60[g]

UV-B dose; 5.4×10^6 erg/cm^2 (2 MED).
a) Casual lipids.
b) Replacement lipids for 2 hr.
c) Mean±S.D. of 10 volunteers.
d) $p<0.01$; significantly different from unirradiated control.
e) $p<0.10$; significantly different from unirradiated control.
f) $p<0.05$; significantly different from unirradiated control.
g) $p<0.01$; significantly different from unirradiated control.

The maximum change was shown after 1 week at the 2 MED irradiated site. It is apparent that TWL values were a maximum after 1 week at all energy dose levels and recovered to unirradiated control values after 3 weeks.

Changes in Skin Surface Lipid Value after UV–B Irradiation

The changes in skin surface lipid value at the 2 MED irradiated site are shown in Table II. Statistical differences from the unirradiated control values were found as follows.

The free cholesterol value in casual lipids increased significantly after 2 weeks ($p<0.01$). The total cholesterol value in casual lipids was also raised significantly after 2 weeks ($p<0.10$). Similarly, significant increases of total cholesterol values in replacement lipids occurred 2 weeks ($p<0.05$) and 3 weeks ($p<0.01$) after UV–B irradiation. To understand these relationships more clearly, the percent change of each lipid value is shown in Fig. 2 and 3.

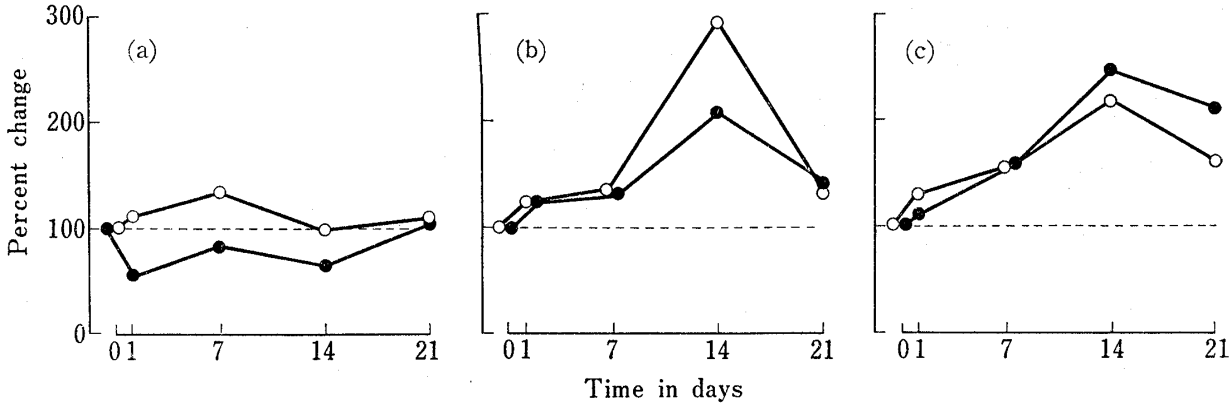

Fig. 3. Time Course of Changes in Squalene, Free- and Total-Cholesterol Values after UV–B Irradiation

The mean percent change was calculated on the basis of the mean value at various days after irradiation.
(a) squalene value; (b) free cholesterol value; (c) total cholesterol value.
—○—: casual lipids, —●—: replacement lipids for 2 hr.
------: unirradiated control level,
UV-B dose: 5.4×10^6 erg/cm² (2 MED).

The total lipid value showed a maximum increase after 1 week and recovered after 2 or 3 weeks to the control level as illustrated in Fig. 2. The squalene value in casual lipids exhibited the maximum increase after 1 week and recovered after 3 weeks to the control level (Fig. 3(a)). On the other hand, the squalene value in replacement lipids showed a decrease until 2 weeks and then recovered to the control level.

The free cholesterol values both in casual and replacement lipids showed the maximum increase after 2 weeks and recovered after 3 weeks to the control level (Fig. 3(b)). Analogous tendencies were observed in the case of total cholesterol values both in casual and replacement lipids as shown in Fig. 3(c). However, the degree of recovery of the total cholesterol value to the control level was less than that of the free cholesterol value.

Discussion

From the results of this experiment, the following considerations concerning the UV–B effect on the skin barrier ability were obtained. TWL values can be regarded as a marker of the *stratum corneum* barrier functions.[4,6]

Maximum erythema was observed after 24 hr at the 1 and 2 MEDs irradiated sites and the erythema gradually faded. The visible erythema is recognized to be due to vasodilation of the subpapillary vascular plexus.[2a,c] In this case, the TWL value showed few changes compared with the control value. Baker *et al.*[7] reported that the TWL value did not increase at the time of erythema formation by vasodilator drugs. From this and the present results, it may be assumed that vasodilation of the subpapillary vascular plexus does not strongly influence the barrier efficacy of the *stratum corneum.*

As shown in Fig. 1, a positive relationship was found between delivered energy and TWL values, especially after 1 week. In addition, it was noteworthy that the TWL value recovered after 3 weeks to the control value demonstrated in Fig. 1. In the case of the maximum TWL increase at 1 week post-exposure, it is suggested that the barrier efficacy of the *stratum corneum* decreased by one fourth compared to the control at the 2 MED irradiated site.

It has already been reported that a dosage of 2 MED of UV–B on human skin causes increased scaling by the 10th day post-exposure.[2c] Microscopic examination shows that

6) *a*) H. Baker, *J. Invest. Dermatol.*, **50**, 283 (1968); *b*) K.J. Ryan and M. Mezei, *J. Pharm. Sci.*, **64**, 671 (1975).
7) H. Baker and A.M. Kligman, *Arch. Derm.*, **96**, 441 (1967).

these scales are caused by parakeratosis-like crusts.[2c] From these results, it is suggested that TWL increase, *i.e.* lowering of barrier functions of the *stratum corneum* is caused by insufficient formation normal morphological *stratum corneum*. Recovery of the TWL value at 3 weeks post-exposure may imply the shedding of the crusts and production of newer normal *stratum corneum*.

Concerning the effect of UV–B on skin lipids, Ohkido *et al.*[8a] reported an increase in cholesterol and Downing *et al.*[8b] reported no significant changes, while Black[8c] reported a decrease.

The present results which demonstrated a significant increase in cholesterol at 2 weeks post-exposure support the results of Ohkido's report.[8a] Increase in cholesterol (*i.e.* epidermal lipid[4,8a,9]) is considered to result from increased horny mass.[2c]

On the contrary, decreased sebaceous lipids, *i.e.* squalene[4,10] might result from a parakeratotic plugging of the sebaceous glands.[2c,8a] The demonstration of no significant differences in replacement lipids may indicate that sebaceous gland activity is not greatly influenced by a single UV–B exposure.

Thus, the pattern of increases in water diffusional loss through the *stratum corneum* and epidermal lipids was considered to be characteristic of UV–B irradiated skin.

The present findings may be useful in clarifying the mechanism of the cause of dry skin after UV–B exposure. In addition, the quantitative changing aspect of skin surface barrier parameters may present a valuable basis for the optimum use and formulation of cosmetological and pharmaceutical products designed for protecting UV-damaged skin.

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