

Abstract No. FO20030042

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Can Daily-use Lipstick Make Lips More Fresh and Healthy? – A New Lipstick Containing α -Glucosyl-hesperidin Can Remove the Dull-color from Lips

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Key words: lipstick, lip dullness, α -glucosyl-hesperidin, blood flow, laser Doppler perfusion imager, confocal microscope

SUMMARY

It has been known that the color of skin reflects the blood flow within. In lips, the capillaries close to the skin surface are numerous; hence lips are redder than the rest of face. However, dermatological research on lips is not as advanced as research on facial or body skin, and little was known about the relationship between relatively dull-colored lips and skin blood flow.

The physiological differences between colorful and dull-colored lips were studied by a two-dimensional laser Doppler blood flow analyzer, a spectrometer for the measurement of the degree of oxygen saturation, and a confocal microscope for observing inside lips non-invasively. Dull-colored lips and the corner of lips (dull-colored compared to the center) showed relatively poor blood flow and lower oxygenated hemoglobin. It was found that colorful lips (generally the young) had a blood flow that tended to run straight in parallel with the skin surface. This unique blood-capillary structure can express clear red blood. Those with dull-colored lips had lost this unique structure. Their blood ran perpendicularly from the deep of the skin and down back again into the deep part as like the blood circulation patterns of facial skin. Therefore, the lips of the latter group had fewer blood capillaries near skin surface in the lips than that of the colorful-lips group. A lipstick containing α -glucosyl-hesperidin, which is derived from certain citrus fruits and can enhance blood circulation, was applied for evaluating its effects. Blood flow was increased 30 min after the application. After two weeks of daily application, the lips' condition became noticeably less dull. These findings suggest that the decrease of blood flow in dull-colored lips is

caused by the loss of the unique capillary structure and the use of the lipstick to increase blood flow can give a vivid color to lips.

INTRODUCTION

The most common lip problem is chapping of lips due to a dry environment. To prevent the problem, the moisturizing lips is the most common solution. Next to chapping, the second problem is the lip color. Although there are many reports regarding studies of skin color especially dullness, there are a few numbers of studies regarding lips due to the unique characteristics of lips, which occupy a small area with an unique shape. One of the reasons why there is a few studies may be due to the fact that the dull-colored lips can be covered by a lipstick. The recent trends in makeup cosmetics are to use lip gloss and pearl glitter giving a juicy and transparent appearance to lips. However, a conventional lipstick giving a transparent appearance is unable to cover up dullness well. Upon the use of the lipstick, the original lipstick color would be affected resulting unexpected lip color. Therefore, physiologic lip color has a major concern among consumers who have notably dull-colored lips. In this report, we studied the blood flow that can control lip color for elucidating the cause of dullness of lips. We also developed a product that can reduce the dullness of lips.

MATERIALS AND METHODS

Generally the lower lip shows greater variation in color compared with the upper lip. In this study, we investigated differences in the physiological parameters of individual volunteer as well as the site on lips. The age of subjects ranged from their twenties to fifties, centering in the twenties to thirties. The dullness of the lower lip was evaluated by three-trained observers who gave a score of 1 – 5 as follows; dull-colored lips, 1 point; slightly dull-colored lips, 2 points; normal lips, 3 points; slightly colorful lips, 4 points; colorful lips 5, points (Table I). The mean value was used as the score of dullness assessed visually for analyzing dermatological parameter measurements.

The lip color parameter was measured using a color analyzer, CR321, MINOLTA, Osaka, Japan. The degree of oxygen saturation of lip blood was measured by a tissue spectrum analyzer, TS 200, SUMITOMO ELECTRIC, Osaka. The two-dimensional blood flow of lip was measured by a laser Doppler perfusion imager, PIMII, Lisca, Linköping, Sweden. The relationship among lip dullness,

lip color, the degree of hemoglobin oxygen saturation, and blood flow was investigated. These parameters were compared between the center of lips and the corner of lips of the individuals who had noticeably colorful lips and those who had relatively dull-colored lips.

The skin beneath the lips was observed with a confocal microscope, Viva Scope™1000, Lucid Technologies, Henrietta, NY, USA. The scope could show the inner structure of the skin.

A lipstick containing α -glucosyl-hesperidin which is derived from the rinds of citrus fruits and known to enhance blood flow when it is used in bath tab, was prepared (Fig. 1). The effect of α -glucosyl-hesperidin on blood flow was also investigated. In addition to the time-course measurement of the blood flow after the application of the lipstick, we also evaluated the lip dullness by visual inspection of trained-investigators after the two weeks of daily application.

RESULTS and DISCUSSION

Comparison of lip dullness at the corner and center of lips

The results of the observation of the lip dullness revealed that there is a significant difference of dullness between the corner of lips and the center of lips (Fig. 2). There was almost no meaningful variation of the dullness scores among three observers on the same subject. This indicates that it is possible to extract the morphological and optical factors for the dullness of lips as reported for dull-colored skin on other parts of body. [1] Spectrometry revealed a significantly lower L^* (the degree of lightness) value for the corner of lips. The a^* (the degree of redness) value was also lower for the corner of lips (Table II). These results supported visual evaluation that the color of the corner of lips is a duller than that of the center of lips.

Thus, since there was a difference in dullness with the site of lips, the following were examined in the individuals as well as in the sites with the different degrees of lip dullness.

Lip dullness and blood flow

The red color of blood that is abundant in the surface of the skin of the lips is responsible for the unique appearance of the lips. Therefore, we studied the differences in blood flow at the various site of lips and between the individuals with various degrees of lip dullness. The results of visual scoring showed that the subjects were categorized into three groups of dullness, that is, those with dull-colored lips ($n = 18$, score: 1 - 2.34), those with the average dullness of lips ($n = 22$, score: 2.35 - 3.67), and those with colorful lips ($n = 6$, score: 3.68 - 5). The blood flow was significantly

lower both at the center (Fig. 3A) and corner of lips (Fig. 3B) of the individuals with dull-colored lips compared with those who have less-dull lips. The blood flow was significantly lower at the corner of the lips that was notably dull-colored (Fig. 4). Therefore, the blood flow appeared to be related to the difference in lip dullness with the individual and with the site. The degree of lip dullness was insignificantly related to age (no data is shown) in this study. However, if more elderly individuals (age: 70 - 80) are entered into the group, there may be found a difference in the dullness of lips with age as seen in skin (e.g. face) except lips.

Lip dullness and the degree of oxygen saturation of hemoglobin

The degree of the oxygen saturation of hemoglobin decreases with the decrease in blood flow and affects the color of the blood. Therefore, we examined the relationship between the lip dullness and the degree of the oxygen saturation of hemoglobin. The results showed that the amount of the oxygenated hemoglobin was significantly decreased in the individuals with notably dull-colored lips (score < 3) compared with those with colorful lips (score \geq 3) (Table III). We also examined the difference in the site. The oxygenated hemoglobin was decreased at the corner of the lips, which were duller in color than that at the center of the lips, which were more colorful (Table IV). Thus, there is a correlation between the dullness and the degree of the oxygen saturation of hemoglobin both with the individual and with the site on the lips.

Observation of dullness and the topography in the lips

Confocal microscope is a useful device for observing the inside of the skin non-invasively. [2, 3] We observed the horizontal section obtained with this confocal microscope and estimated the topography in the skin. Generally in the horizontal section, the dermis can be observed as a circle shape surrounded with the epidermis. Generally, the epidermal-dermal junction shows a structure resembling a wavy structure (Fig. 5). Using this confocal microscope, the relationship between the dullness of lips and the topography in the skin was investigated. In relatively colorful lips (generally the young), the blood vessels were straight and ran in parallel to the skin surface in the papilla of the dermis (Fig. 6A). That is, in the colorful lips, the epidermal-dermal junction had a wavy structure in parallel to the skin surface (Fig. 7A). The lips showed a unique topography with the blood vessels in parallel to the epidermis at the dermis (relatively near the skin surface) protruding into the epidermis, which clearly shows the redness of the blood appearing on the lips. This unique topography has been unable to be observed in other skin site. On the other hand, in

dull-colored lips (generally the elderly), the characteristic structure was absent, and the papilla in the dermis showed an atypical structure (Fig. 6B). There was no dermal space in which the blood vessels should be arranged in parallel to the skin surface, and the blood vessels that run near the skin surface immediately sunk deeply (Fig. 7B). There were few capillary blood vessels near the surface of the skin, and it was difficult to recognize the red color of the blood from the skin surface. In the present study, the sufficient observations that can unveil the details of the change in the structures within the lips in relation to aging or dullness were unavailable. Therefore, the future study that can investigate various factors affecting the arrangement of the blood vessels in the lips in relation between the dullness of lips and the topography in the lips will be necessary. Our attempt to re-produce a 3D structure from the cross-sections was unsuccessful, because a slight perpendicular movement of the lips caused by the heartbeat could give no stable continuous cross-section series. [4, 5]

The effect of enhanced circulation on the dullness of lips

From ancient times, citrus fruit have been known to promote the effect of bathing. [6] A component in these citrus fruits, hesperidin, is known to strengthen capillary blood vessels, suppress the permeability of blood vessels, and reduce blood pressure. In Europe, it is used orally for chronic vascular diseases. A lipstick containing 0.5% α -glucosyl-hesperidin, which was a water-soluble form of hesperidin made by glucosylation was prepared for investigating the effect of the compound using 37 panels (Fig. 1). [7] The panels were randomly divided into two groups; one group would use a lipstick that contained α -glucosyl-hesperidin added, and other would use the control lipstick. In the lips of the treated group, the blood flow was found to increase at both the corner and the center of lips from 30 min after the application, and the effect continued up to 90 min (Fig. 8). There was no significant difference between the two groups in either the dullness score or blood flow before the application (no data is shown). Although the blood flow in the corner of lips increased at 30 min after the application even in the control group. The degree of the increase of blood flow in the applying lipstick with α -glucosyl-hesperidin was higher than that of the control. This result indicates that in addition to the massage effect (a rubbing effect - one of physical effect) of the application on the blood flow, α -glucosyl-hesperidin can increase the blood flow. The dullness scores before and after the two-weeks daily application of a lipstick containing α -glucosyl-hesperidin were also compared. Although the dullness score showed an improvement

both with and without α -glucosyl-hesperidin, there was a significant improvement in the group using the lipstick containing α -glucosyl-hesperidin (Table V). The dullness of skin is thought to be affected by the amount of hemoglobin and melanin, and the morphology of horny surface. [1] No studies regarding the effects of melanin and the morphology on the dullness of lips was performed in the present study resulting that their roles remain unknown. However, since the application of α -glucosyl-hesperidin which increased the blood flow in the lips improved the dullness of the lips, it is suggested that the decrease in blood flow in the lips due to various factors is a major cause of dullness and plays an especially large role in determining the color of lips.

Various studies have been done on the dullness of skin. [8] Furthermore, studies have been done on the lips as a site that is easy to dry. [9] However, a few physiological studies have been done on the dullness of lips. Although the mechanism underlying the improvement of dullness by the increased blood flow still remains unknown, the present studies showed at the first time that the dullness of the lips is largely related to the decrease in blood flow.

CONCLUSIONS

In the notably dull-colored lips, the L^* value, a^* value, the blood flow, and the oxygenated tissue hemoglobin were lowered. The observations by confocal microscopy revealed changes in the unique topography at the epidermal-dermal junction and the arrangement of the blood vessels as a cause of the dullness of lips. The dullness of lips was improved by the repeated application of lipstick containing α -glucosyl-hesperidin. These findings show that (1) a relationship between the dullness of lips and the decrease in blood flow, and that (2) the daily use of lipstick to increase blood flow can give a vivid color to lips.

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Table I. The lip dullness evaluation score

.....Score	
Dull-colored	1
Slightly dull-colored	2
Normal	3
Slightly colorful	4
Colorful	5

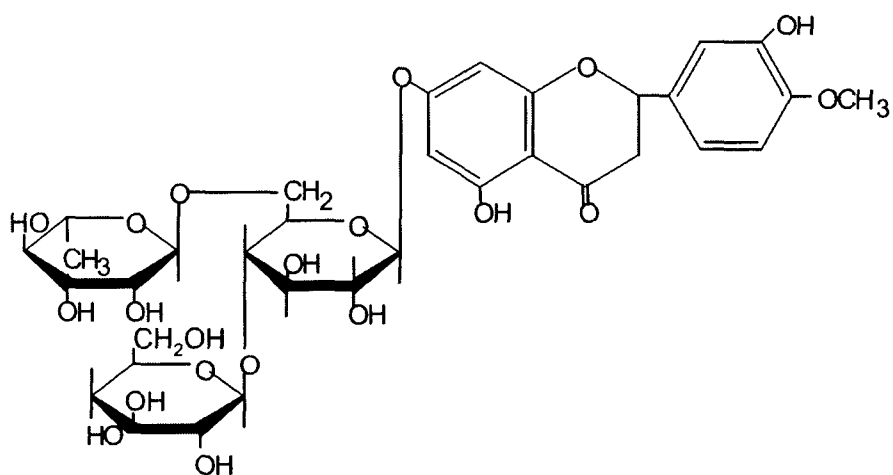


Fig. 1 Structural formula of α -glucosyl-hesperidin

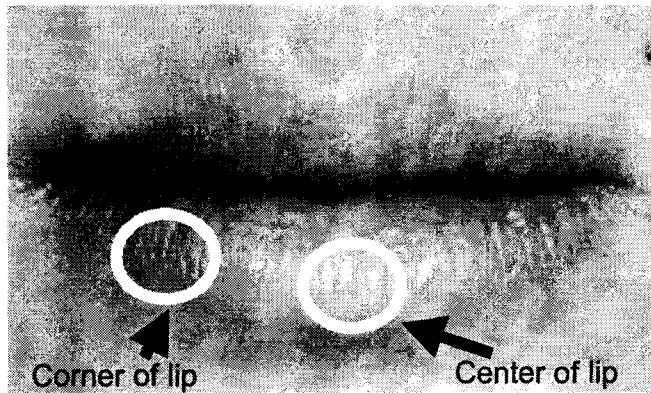


Fig. 2 The site of lips color evaluation. The mean score and the standard deviation of the lip dullness of the center and the corner were 2.83 ± 0.64 and 1.94 ± 0.23 , respectively. There is a statistical significance. (n = 46, p < 0.05, Wilcoxon)

Table II. Difference in color values between the center of lips and the corner of lips

Color value	Center	Corner
L*	49.45 ± 2.28	48.77 ± 2.22
a*	14.21 ± 1.97	13.58 ± 1.57

L* and a* represent the degree of lightness and redness, respectively.
 * p < 0.05 (n = 46, paired t-test)

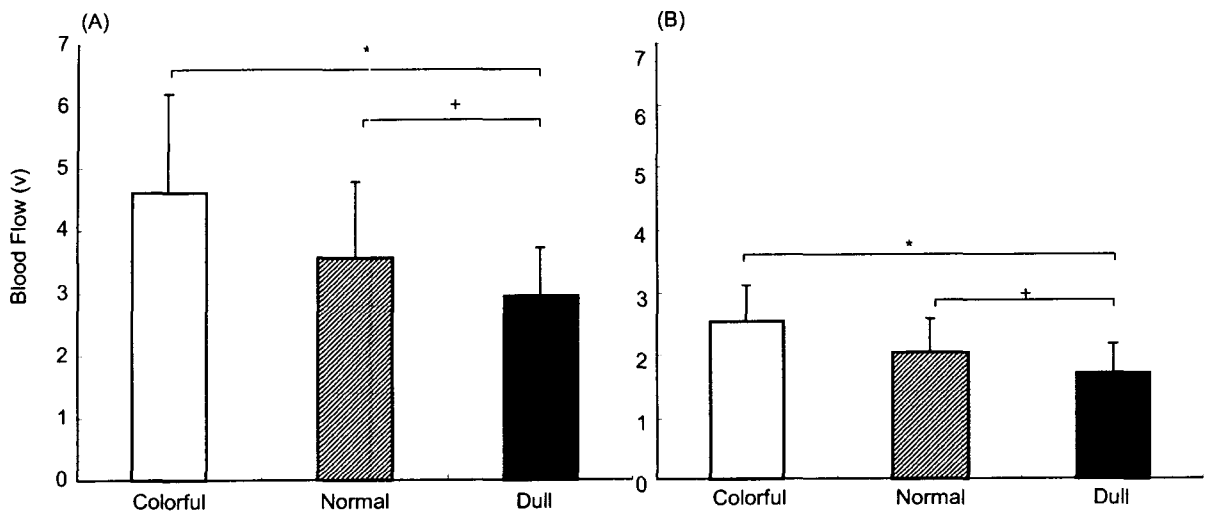


Fig. 3 Difference in blood flow between subjects who have noticeably colorful lips and those whose lips are relatively dull. (A) Blood flow measured at the center of lips; (B) blood flow measured at the corner of lips. The columns and bars represent the mean value and the standard deviation, respectively. (n = 46, * p < 0.05, + p < 0.1)

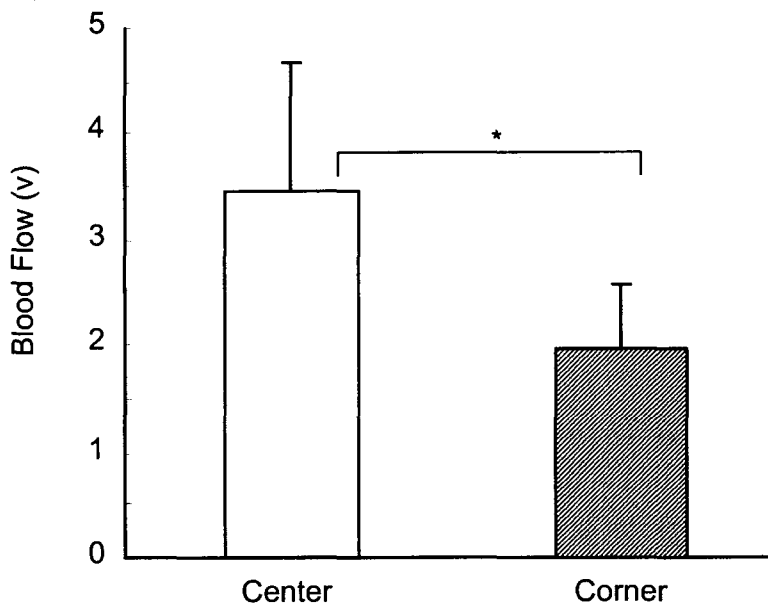


Fig. 4 Difference in blood flow between the center of lips and the corner of lips. The columns and bars represent the mean value and the standard deviation, respectively. (n = 46, * p < 0.05, paired t-test)

Table III. The difference in the degree of oxygen saturation of hemoglobin in colorful and dull lips

The site of lips	Colorful	Dull
Center	42.47 ± 3.76	36.18 ± 3.50
Corner	41.94 ± 4.84	33.93 ± 4.36

* p < 0.05, n = 14

Table IV. Difference in the degree of oxygen saturation of hemoglobin between the center of lips and the corner of lips

Center	Corner
41.34 ± 4.84	39.14 ± 5.94

* p < 0.05, (n = 14, paired t-test)

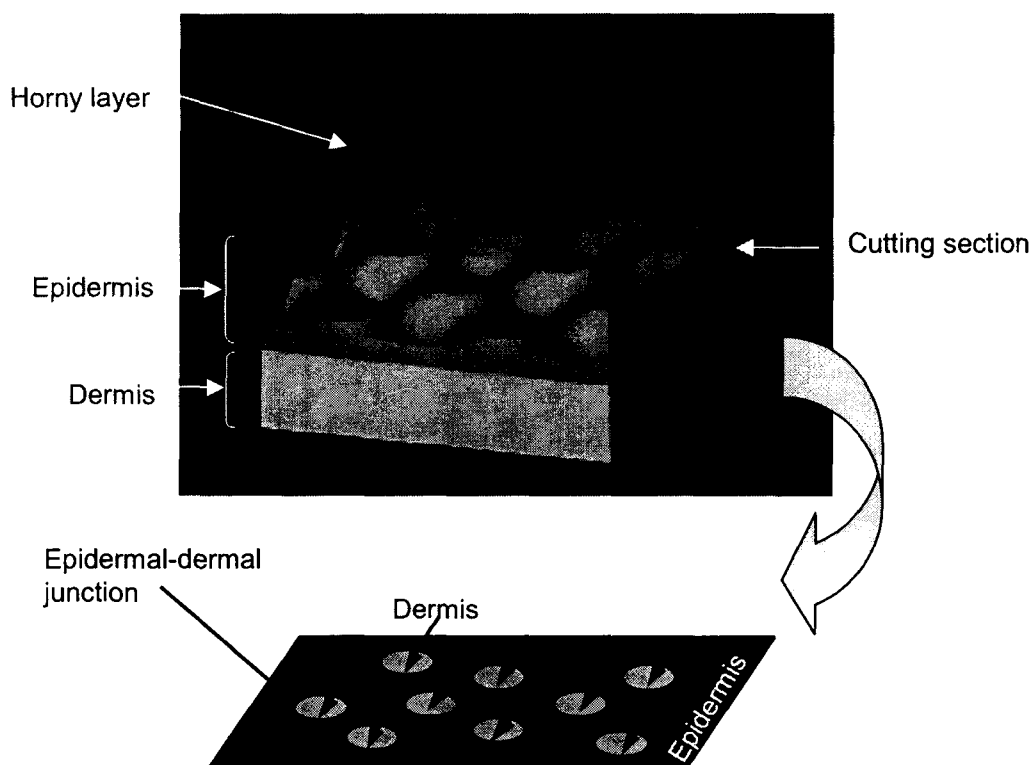


Fig. 5 Epidermal-dermal junction model. Horizontal section of lips can be observed using a *in vivo* confocal microscope

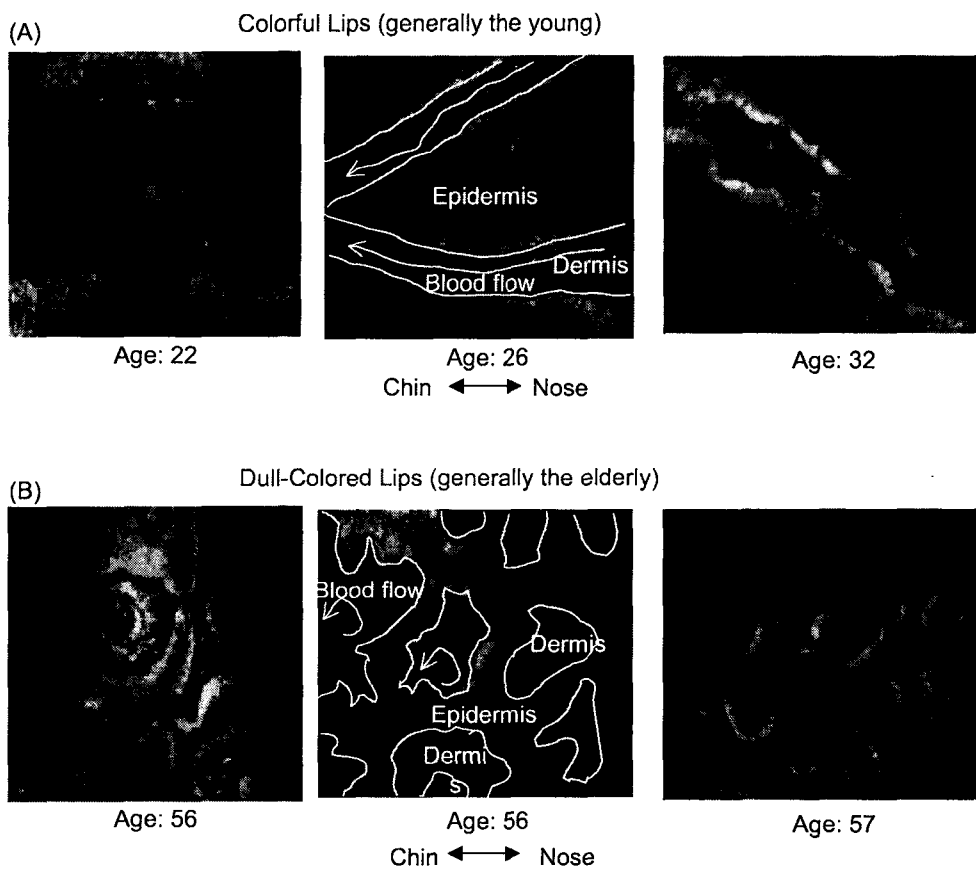


Fig. 6 Topography of the epidermal-dermal junction of lips. (A) Unique topography was obtained by a *in vivo* confocal microscope in colorful lips. (B) No unique topography was observed in dull-colored lips.

(A) Colorful Lips (generally the young)



(B) Dull-Colored Lips (generally the elderly)

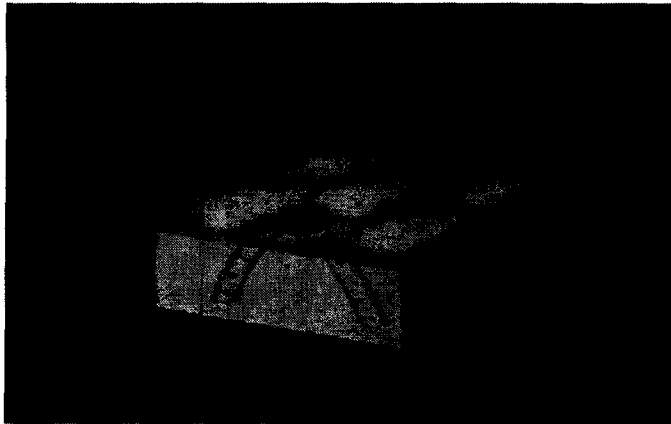


Fig. 7 Schematic illustration of topography of the epidermal-dermal junction and the arrangement of blood vessels of lips. These are made by a computer based on the topographies shown on Fig. 6. (A) There is unique topography in colorful lips. (B) There is no unique topography in dull-colored lips.

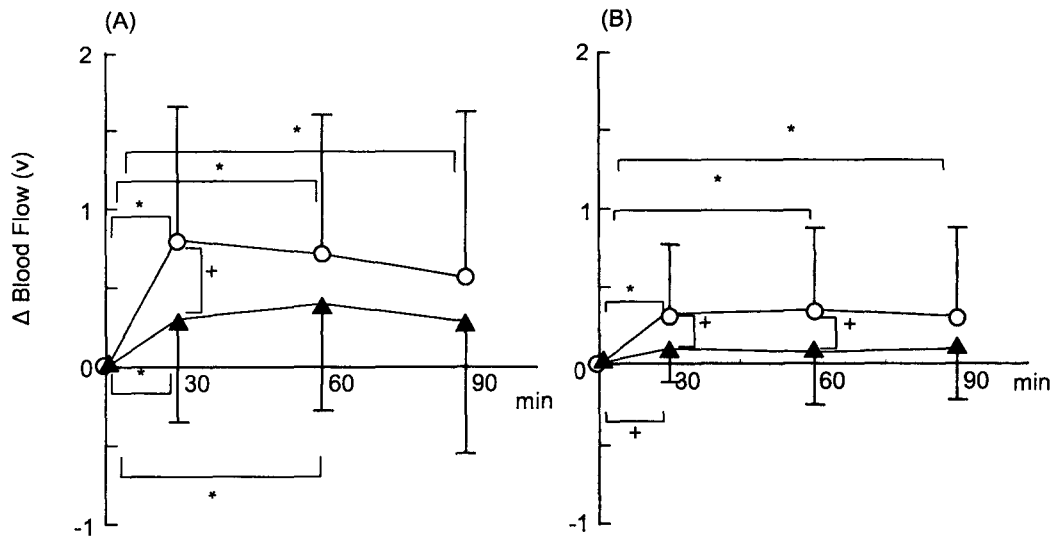


Fig. 8 Increase of blood flow after application of lipstick containing 0.5% α -glucosyl-hesperidin. The Δ blood flow means the difference of blood flow before and after treatment. (A) Δ blood flow measured at the center of lips; (B) Δ blood flow measured at the corner of lips. The open circle and closed triangle represent Δ blood flow of lips treated with lipstick containing 0.5% α -glucosyl-hesperidin and the control lipstick, respectively. (n = 37, * p < 0.05, + p < 0.1)

Table V. The effect of the daily application of a lipstick containing α -glucosyl-hesperidin on the dullness of lips (two weeks)

The site of lips	Control	α -glucosyl-hesperidin
Center	0.41 \pm 0.46	0.87 \pm 0.33
Corner	0.14 \pm 0.57	0.65 \pm 0.37

* p < 0.05, (n = 37, Wilcoxon test)