Maximal Tension of Human Epidermis Prepared from Suction Blisters

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Background: Epidermis is directly exposed to various external stimuli and protects internal structures. Most studies about skin tension focused on the dermis, and epidermal tension has not been able to attract interest because dermis much thicker than epidermis and dermal collagen fibers play a predominant role in the skin tension. However, circumstances which involve only the epidermis are often shown, and thus it is necessary to study the mechanical property of the epidermis.

Objective: The purpose of this study is to quantify the role of the epidermis as a mechanical barrier.

Methods: Human epidermal sheets were obtained from suction blisters in 14 patients with vitiligo during epidermal grafting. Maximal tension, that is the power required to break the epidermal sheets, was measured by tensiometer.

Results: The maximal tension of the epidermis ranged from 40.0 g/cm² to 84.5 g/cm² with a mean values of 56.1 g/cm² and did not differ significantly according to the age or anatomical sites.

Conclusion: The barrier function of epidermis is important and the results are expected to be used as the basic information for other studies about the mechanical property of the epidermis. In addition, this will be important data in developing an artificial skin whose mechanical property is similar to that of the human skin.


Key Words: Epidermis, Suction blister, Tension

Although skin has natural tension even at rest, it is loaded by tissue edema or tumors internally and by external mechanical forces. There have been many studies about natural skin tension and the effects of skin tension on postoperative scars. However, these studies focused mainly on changes of dermal collagen fibers, and studies on the epidermis alone are rare.

In a study on the epidermis alone, Wildnauer et al.1 measured breaking strength using epidermis prepared from cantharidin-induced blisters and observed the relationship between the water content of the epidermis and the breaking strength. Squier2 reported that the stretched skin showed a hyperplastic response of the epidermis with a thickening of all cell layers which was due to the increased mitotic activity of the epidermis.

In this study, in order to assess the mechanical barrier functions of the epidermis, maximal tensions required for breaking epidermis were measured using epidermis prepared from suction blisters.

MATERIALS AND METHODS

Subjects

Fourteen patients with vitiligo were selected. They consisted of 1 male and 13 females, and

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Table 1. Maximal tension of epidermis

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Age/Sex</th>
<th>Site</th>
<th>Tension (g/cm²)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>13/M</td>
<td>Chest</td>
<td>48.7</td>
</tr>
<tr>
<td>2</td>
<td>15/F</td>
<td>Chest</td>
<td>40.0</td>
</tr>
<tr>
<td>3</td>
<td>16/F</td>
<td>Chest</td>
<td>47.4</td>
</tr>
<tr>
<td>4</td>
<td>19/F</td>
<td>Chest</td>
<td>65.6</td>
</tr>
<tr>
<td>5</td>
<td>19/F</td>
<td>Chest</td>
<td>59.1</td>
</tr>
<tr>
<td>6</td>
<td>20/F</td>
<td>Abdomen</td>
<td>51.3</td>
</tr>
<tr>
<td>7</td>
<td>20/F</td>
<td>Abdomen</td>
<td>46.8</td>
</tr>
<tr>
<td>8</td>
<td>22/F</td>
<td>Chest</td>
<td>84.5</td>
</tr>
<tr>
<td>9</td>
<td>29/F</td>
<td>Chest</td>
<td>63.0</td>
</tr>
<tr>
<td>10</td>
<td>31/F</td>
<td>Chest</td>
<td>57.8</td>
</tr>
<tr>
<td>11</td>
<td>37/F</td>
<td>Abdomen</td>
<td>46.8</td>
</tr>
<tr>
<td>12</td>
<td>37/F</td>
<td>Abdomen</td>
<td>53.9</td>
</tr>
<tr>
<td>13</td>
<td>58/F</td>
<td>Back</td>
<td>66.9</td>
</tr>
<tr>
<td>14</td>
<td>59/F</td>
<td>Back</td>
<td>53.9</td>
</tr>
</tbody>
</table>

Mean ± S.D. 56.1 ± 11.3

their ages ranged from 13 to 59 with a mean values of 28.2 (Table 1).

Formation of suction blister
The donor site was prepared with betadine and alcohol sponges. Five milliliters of 1% lidocaine was injected intradermally to make the indurations of 2.5-3cm in diameter for easy blistering. Bell-shaped glass suction tips were connected with Electric suction unit (CHS-708, Choongwae machinery Co., Korea) by rubber tubes. The suction tips were fixed on the skin surface around 200-400mmHg. After about 3 hours, large bullae, 3cm in diameter appeared (Fig. 1).

Obtaining epidermal sheet from suction blister
The roof of the bulla was separated by snipping around the periphery and it was spread over the surface of a glass syringe with the horny layer directed against the surface of the syringe. The basement membrane and fibrin clot were removed from epidermal sheet. The glass syringe with the epidermal sheet was rolled over the inner part of the device fixing the epidermis.

Measurement of tension
After the outer cover of the device was placed on the epidermal sheet, a sensor of digital force gauge (PDE-2S, Attonic, Japan) was applied perpendicularly to the epidermal sheet through the central hole of the device (Fig. 2) until the epidermal sheet was broken. The maximal tension was checked at the time of breaking.

Statistical analysis
The results were analyzed with the Kruskal-Wallis test using a PC-SAS package and considered as significance when the p value was lower than 0.05.

RESULTS

Sites of suction blister were the chest, abdomen, and back and the number of each site was 8, 4, and 2.

Measured maximal tension was 56.1g/cm² totally and 58.3g/cm² in the chest, 49.7g/cm² in the abdomen, and 60.4g/cm² in the back. Maximal tension did not differ significantly by the anatomic sites (Table 1, 2).
Table 2. Comparison of maximal tension of epidermis according to the site

<table>
<thead>
<tr>
<th>Site</th>
<th>No.</th>
<th>Tension (g/cm²)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chest</td>
<td>8</td>
<td>58.3 ± 13.7</td>
</tr>
<tr>
<td>Abdomen</td>
<td>4</td>
<td>40.7 ± 3.5</td>
</tr>
<tr>
<td>Back</td>
<td>2</td>
<td>60.4 ± 9.1</td>
</tr>
</tbody>
</table>

p>0.05; statistically insignificant

Table 3. Comparison of maximal tension of epidermis according to age

<table>
<thead>
<tr>
<th>Age</th>
<th>No.</th>
<th>Tension (g/cm²)</th>
</tr>
</thead>
<tbody>
<tr>
<td>13 - 19</td>
<td>5</td>
<td>52.3 ± 9.9</td>
</tr>
<tr>
<td>20 - 29</td>
<td>4</td>
<td>61.4 ± 16.8</td>
</tr>
<tr>
<td>30 - 59</td>
<td>5</td>
<td>56.4 ± 8.4</td>
</tr>
</tbody>
</table>

p>0.05; statistically insignificant

When comparing the maximal tension of the epidermis by the age groups, teenagers showed 52.3g/cm² and the age of 20-29 and more than 30 showed 61.4 and 56.4g/cm² respectively. There were no significant differences among the groups (Table 3).

DISCUSSION

Skin tension causes a maximal tension line on the skin and the maximal tension line is important in incising the skin in dermatologic surgery. Skin tension should be considered when the wound is sutured because the width of scar is proportional to skin tension. In addition, it is expected that the monitoring of mechanical properties including skin tension can become a useful means of assessing structural changes resulting from either therapy or pathology.

In 1965, Gibson et al. observed that skin was extended through three phases by loading. During the initial phase of extension at low load, there was a fairly rapid increase in length for very small increments of force. Terminally, only minor degrees of extension resulted from relatively large increases in force. Between these two portions, there was a transition phase. Normal physiological activity caused strains in the first half of the second phase and extension of the skin beyond the third phase initiated rupture of the skin.

Histologic changes of the epidermis resulted from the extension of the skin. The epidermis has a two-stage response to progressive straining. Initially, rete ridges are flattened. Further extension of the skin elongates the cells themselves. The strain at which cell elongation begins can vary from near the end of phase 1 to the middle of phase 2 of extension.

There are two tensions in vivo human skin, static and dynamic tension. The static tensions are the natural tensions existing in the skin. Clinical evidence of static tensions is that the edges of an excised wound retract and pieces of excised human skin contract after removal from the body. Alexander and Cook determined the natural tension using a strain gauged pretension device and suggested that mechanical properties characterization would be a useful tool in the evaluation of the severity of certain pathologic states and the effect of therapy. Dynamic tensions are caused by a combination of forces which are associated with joint movement, mimetic and other voluntary muscle activity, and gravity.

There have been various animal experiments studying the breaking strength of skin. In the Beauchene et al. study using an animal model, a Silastic expander was inserted into the peritoneal cavity. The breaking strength was consistent and independent of duration of expansion.

Most studies mentioned above were in vivo studies on skin including both epidermis and dermis, so that they did not exclude the factors influencing the skin tension such as subcutaneous fat or fascia. In vitro animal experiments also included both epidermis and dermis.

An in vitro study on human epidermis alone was performed by Wildnauer et al. in 1970. They measured the breaking strength of the epidermis prepared from a cantharidin-induced blister and observed that the breaking strength of epidermis was reduced by an increase of relative humidity. Also they observed with a scanning electron microscopy that the epidermis was broken at an intercellular junction.

Other methods separating the dermo-epidermal junction are enzymes such as papain or trypsin, cold, or suction. Papain or trypsin is a strong proteolytic enzyme. It induces acantholysis by the degradative effect of the enzyme on the protein-
polysaccharide matrix which joins together the various elements of the desmosome. Therefore, the preparation of the epidermis with this enzyme is not suitable for the measurement of maximal tension of epidermis. Cold-induced blisters are formed by the separation of the basement membrane zone. The architecture of the cell is kept normal, but keratinocytes exhibit intracellular edema and damage to the hemidesmosome, and amorphous and fibrillar materials are deposited within and beneath the basal lamina. In fact, we observed that the epidermis prepared from a cold-induced blister showed thicker and more rigid than normal epidermis, so that it was expected that measured maximal tension in a cold-induced blister was higher than that in normal epidermis.

In this study, maximal tension was measured in the epidermis prepared from a suction blister. A suction blister is a method of obtaining the epidermis for donation in an epidermal graft, which is a surgical treatment for depigmentation of skin caused by various etiologies. Histologic changes in a suction blister have been noted. Separation develops at the plane between the basement membrane and basal layer. Suvanprakorn et al. demonstrated that separation developed in the lamina lucida using bullous pemphigoid antibody. The microscopic vesicles appear first on the tips of the dermal papillae and the basal layer of the detached epidermis retained its normal columnar pattern and unwidened intercellular spaces. In contrast, the periadnexal portion where the epidermis is attached firmly to the dermis shows cellular stretching and widening of desmosome. On the electron microscopy, suction does not cause an alteration of the shape of the cell and intracellular organelles, but causes the crescentic displacement of nuclei and paranuclear vacuolization. Therefore the epidermis prepared from a suction blister keeps the similar state to normal epidermis. In particular, the desmosomes are intact, so that measured maximal tension using epidermis prepared from a suction blister enables maximal tension of normal epidermis to be predicted. However, it is expected that measured maximal tension of epidermis prepared from a suction blister is lower than that of normal epidermis, because the epidermis has already been pulled during the formation of the suction blister.

Although there is a loss of dermal thickness resulting from the degeneration of dermal fibers with aging, the average epidermal thickness appears constant with increasing age. Thus, the maximal tension of epidermis is expected to be independent of age, and our results confirmed this. The thickness of the epidermis varies by site. The thinnest region is the upper eyelid and the thickest regions are the palms and soles. Therefore, where the epidermis is thicker, such as on the palms and soles, it is likely to have a significant effect on the skin's mechanical properties. Because our study did not include these areas, there were no significant differences between sites.

The barrier function of the epidermis is important and we would like to assess the epidermis as a mechanical barrier. The results above are expected to be used as the basic information for other studies regarding the mechanical property of the epidermis. In addition, this will be important data in developing an artificial skin whose mechanical property is similar to that of the human skin.

REFERENCES

7. Beauchene JG, Chambers MM, Pearson AE, Scott PG: Biochemical, biomechanical, and physical changes in the skin in an experimen-