

Epidermal Skin Barrier

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Abstract

Stratum corneum made from cell division and differentiation of epidermal keratinocytes has been identified as a core tissue in the skin barrier. In the stratum corneum formation stage, abnormality and imbalances at each step cause functional disorder of skin barrier. In order to better understand the factors that regulate functions of the stratum corneum, this paper described the components of the skin barrier in detail. From the description of "Brick & Mortar", the major components of the skin barrier such as lamellar bodies (LB), cornified cell envelope (CE), lipid, calcium, cytokine and moisturizer were described in detail. Two types of granules are formed epidermal keratinocyte on the stratum granulosum, including keratohyalin granule filled with protein and lamellar bodies filled with lipid, and keratin protein membrane and keratin lipid membrane together are called CE which forms the boundary of epidermal keratinocyte. Filaggrin acts as an adhesive to adhere keratin together, and is also involved in the binding of protein-forming CE. Skin lipids play an important role with their barrier function, and can also bring various effects on the skin as signaling molecules in addition to supporting its defense mechanism such as antibacterial function. Calcium is involved in the structure-retention of LB and secretion of LB to intercellular spaces, and calcium gradient plays an important role in the formation of a skin barrier. When the skin barrier is damaged, substances called cytokines such as tumor necrosis factor (TNF), interleukin 1 (IL1) and interleukin 6 (IL6), are secreted. However, if the barrier damage persists and these cytokines increase chronically, it may cause an adverse result such as inflammation or cuticularization. The concept of moisturizer in the past was understood as the replenishment of moisture to skin, but recently attention has been focused on the restoration of the barrier function, which is the most important function of skin, rather than the simple replenishment of moisture. Currently, physiologic lipid mixture can be considered as the most ideal moisturizer.

Keywords: Skin barrier, Lamellar body, Filaggrin, Cornified cell envelope, Moisturizer

Introduction

Skin is a dynamic organ which creates epidermis over one's lifetime through cell division and differentiation. Stratum corneum has been identified as a core tissue of the skin barrier that can withstand physical and chemical damages as well (Oh & Jang, 2015). The skin barrier function of the epidermis enables normal biochemical metabolisms, and protects skin from drying and external harmful factors (Chang & Lee, 2012).

Epidermal keratinocyte creates various proteins and lipids through the cell division and differentiation from epidermal stem cells on stratum basale, and finally forms stratum corneum via this phenomenon to rapidly change into corneocyte (Hong, 2011; Oh & Jang, 2015; Yoon *et al.*, 2013).

Stratum corneum structure consists of corneocyte, cornified cell envelope, intercorneocyte lipid (lamellar membrane lipid) and corneodesmosome, and it serves as the skin barrier (Youn, 2013). When the differentiation process of epidermal

keratinocyte is carried out abnormally, functional disorders of the skin barrier arise (Oh & Jang, 2015). The skin barrier can be divided into 4 types as physical, chemical, biochemical and immunological barrier (Jin & Lee, 2014), and stratum corneum has been reported as having an influence on simple barrier functions as well as the structure and the functional roles of living cell layers such as epidermis and dermis (Jeong *et al.*, 2009). Skin barrier research recently has been emerging as the most popular field of study in the dermatology and cosmetics industry due to its aesthetic importance (Kim & Jeong, 2012; Lee, 2014).

And to conclude, the epidermal keratinocytes form the stratum corneum via the proliferation, differentiation and a sort of apoptosis. These finally go through the desquamation process. Therefore, it is essential to understand the mechanism and factors that control these processes (Kim & Lee, 2008), and the main components of skin barrier are examined as follows.

Components of the skin barrier

1. Bricks and Mortar model

Stratum corneum consists of corneocytes that cross-linked with lipids and proteins (Park *et al.*, 2001). Epidermal keratinocyte changes to corneocyte through proliferation and final differentiation, and it desquamates as a result of protease existing in the stratum corneum, a process that generates various proteins and lipids. The most typical model used to explain the structure and function of the stratum corneum is the two-compartment model, which was explained as a “Brick & Mortar” model by Elias (1996). This model explains stratum corneum consisting of protein (40%), lipid and moisture (20%) as the bricks and intercorneocyte lipid as the mortar, and corneodesmosome which is the protein structure connecting corneocytes, cornified envelope and cornified lipid envelope surrounding corneocytes are representative components of stratum corneum, which prevent fluids in the body from being lost and block the penetration of harmful substances from the outside (Kim & Jeong, 2012).

2. Lamellar bodies, profilaggrin/filaggrin

Lamellar bodies (LB) formed on Golgi apparatus include lipid components that will form intercorneocyte

lipid, various lysosomal enzymes that provide the catalytic action for hydrolysis of corneodesmosome, lipids including phospholipids, sphingomyelin, glucosylceramide and cholesterol and enzymes including lipid hydrolase, lysozyme, protease and acid phosphatase, helping to form the barrier function (Hong, 2011). LB starts to appear from the top of the stratum spinosum and it exists most abundantly in the stratum granulosum, adhering to cell envelope on the boundary between stratum granulosum and stratum corneum and discharging lipids to intercellular space through extracellular secretion. ω -Hydroxyceramides are included inside LB of corneocyte lipid envelope (Im, 2014).

When epidermal keratinocyte reaches the stratum granulosum, it synthesizes keratohyalin granules which contain profilaggrin and loricrin. In normal skin, filaggrin exists inside keratohyalin granules in form of profilaggrin, which is a precursor and is hydrolyzed by proteolytic enzymes and becomes filaggrin through the dephosphorylation process. As the name suggests, a filament aggregating protein (filaggrin) acts as an adhesive to glue keratins together and it is also involved in binding of proteins composing cornified cell envelope such as involucrin and loricrin. Actually, its correlation in that the defect of filaggrin gene was observed from patients with atopic dermatitis and reduced ceramide on the lipid envelope of barrier has been reported, and trans-urocanic acid which is the final metabolite of this protein and some amino acids (arginine, glutamine and histidine) also act as natural moisturizing factor (NMF) which has the function of adjusting the pH of stratum corneum and protecting skin from UV rays and role of humectants (Kim, 2008; Kim & Jeong, 2012).

3. Lamellar membrane lipid, intercorneocyte lipid

In sensitive, dry and atopic skin, it is known that the function of lipid envelope which is the skin barrier is often damaged (Chang & Lee, 2012). Generally, skin lipids which act as barriers refer to intercorneocyte lipid and originate from LB. Intercorneocyte lipid is considered the mortar in the ‘Brick and Mortar model’ and it accounts for approximately 10% of the mass of stratum corneum (Im, 2014). Skin barrier lipid consists of ceramide, cholesterol and free fatty acids at a molecular ratio of 1:1:1, and the permeability barrier and antimicrobial barrier of stratum corneum coexist. Sphingolipids which are typical skin barrier lipids

and sphingolipid metabolite sphingosine 1-phosphate also provide the antimicrobial action (Kim, 2008).

It is considered that a functional change of skin barrier in aged skin results from abnormality in important lipid components for forming the lamellar structure of stratum corneum and the function of skin barrier deteriorates accordingly (Park *et al.*, 2001), and important elements (moisture content of stratum corneum, epidermis pH) for maintenance and the basic functions of skin barrier are damaged and lipid synthesis is reduced so that total lipid content of intercorneocyte lipid envelope decreases (Choi & Park, 2010). Intercorneocyte lipid forms lipid envelope in various layers called a lamellar structure, and in particular, the ceramide element is known to play an important role in the function of intercorneocyte lipid, and various moisturizers such as physiologic lipid mixture are being developed based on this premise (Choi, 2001; Jeong *et al.*, 2009).

4. Cornified cell envelope (CE)

Two types of granules are formed inside epidermal keratinocyte on the stratum granulosum including keratohyalin filled with protein and lamellar bodies filled with lipid, and cornified protein envelope and cornified lipid envelope together are called cornified cell envelope (CE), which forms the boundary of corneocytes. In the beginning of the process of generating CE, envoplakin and involucrin adhere to the cell plasma membrane. Loricrin secreted from keratohyaline granule of the stratum granulosum is deposited on the desmosome of the cell envelope, and filaggrin aggregates keratin in the cell, forming macrofibril. The cornified lipid envelope is the lipid envelope generated when proteins forming the cornified envelope based on involucrin existing in corneocyte are connected to ω -hydroxyceramides among the lipids outside the corneocyte through covalent bonding. CE is an insoluble composite protein and lipid envelope (approximately 10 nm) and it has the structure of a protein envelope inside and a lipid envelope outside. As the protein envelope is differentiated and the calcium concentration inside the epidermal keratinocyte increases, the expression of envoplakin and periplakin occurs in the corneodesmosome and involucrin is located near the cell envelope. Involucrin is combined through cross-linkage between involucrins or involucrin, envoplakin and periplakin by membrane-bound transglutaminase 1 (TGM1) and such desmosome surrounds

along the inner surface of the cell envelope including corneodesmosome, forming a layer. In this process, loricrin, small proline rich proteins (SPRRs) and elafin are also mixed and combined with the protein envelope as well as keratin fibers so that the envelope protein is connected to the inside of the cell. Loricrin is combined with SPRRs by transglutaminase-3 more inside than involucrin, becoming a major component which accounts for 80% of the cornified cell envelope (Hong, 2011; Kalinin *et al.*, 2001; Kim & Jeong, 2012).

5. Epidermal calcium gradient, cytokine

Calcium is an essential element for controlling the homeostasis of the skin barrier, and a change in the concentration of the calcium ion on the epidermis is known to be an important control factor for recovery after skin barrier damage. The decrease of the calcium concentration acts as a signal to secrete LB and synthesize lipids, and this supports the premise that calcium gradient plays an important role in forming the skin barrier. However, it is still not clear whether or not the development of a skin barrier is induced by a difference in calcium concentration in epidermis, or whether or not the skin barrier causes a difference in the calcium concentration in the epidermis (Kim, 2010; Ko, 2014). When the skin barrier is under damaged circumstance, cytokines, such as tumor necrosis factor (TNF) and interleukin (IL), especially IL1 and IL6, are secreted at first. However, if the barrier damage persists and these cytokines rise chronically, it may cause an adverse result such as inflammation or proliferation (Jin & Lee, 2014).

According to the skin dryness process, a "cytokine cascade" is formed due to changes in the moisture inflow, overall ion distribution and secretion of various cytokines such as IL1 on the stratum corneum during the epidermal damage process. Also, as DNA synthesis increases, abnormal corneocyte proliferation occurs, causing inflammation and the barrier restoration such as secretion of lamellar body or proliferation of biosynthesis of lipids also occurs due to epidermal barrier damage. Abnormal epidermal keratinocyte differentiation occurs due to cytokine cascade and scale occurs due to desquamation of damaged stratum corneum. This leads to a vicious circle of transepidermal water loss (TEWL) increase, decrease in moisture content of stratum corneum, loss of epidermis NMF and enzyme denaturation of stratum

corneum, making skin dryness worse (Kim, 2007).

One controversial issue is the question of whether or not skin barrier damage is involved in increasing the sensitization of a child with atopic dermatitis to allergens. If skin barrier damage is involved in the sensitization to respiratory allergens, it can be said that skin barrier damage plays a key role in the process by which atopic dermatitis develops into asthma. An interesting point is the fact that IL4, IL13 and IL25 reduces keratinocyte's filaggrin synthesis, and this becomes important evidence for presenting the model that skin barrier damage induces an inflammatory response and the induced inflammatory response damages the skin barrier again (Cho, 2012).

6. Moisturizer

Stratum corneum is the most important structure for skin moisture, and the epidermal permeability barrier is closely related to water content since the movement of moisture inside and outside the skin is controlled through the stratum corneum and a moisturizing factor which can draw and contain moisture exists (Hong, 2014). Elements generally known to be related to moisture content on stratum corneum include intercorneocyte lipid envelope, natural moisturizing factor and glycerol on stratum corneum (Choi & Park, 2010).

When the barrier function of skin is damaged, the maintenance of homeostasis such as lipid secretion and synthesis occurs, and it is known that when an external ointment containing intercorneocyte lipid component is applied to skin with a damaged skin barrier function, the skin's barrier function is restored (Jeong *et al.*, 2009).

The biggest benefit that cosmetic products can offer to the skin barrier function is the moisturizing effect. The skin barrier is the most influential part for cosmetic products (Nam *et al.*, 2012). Recently, more attention has been focused on the restoration of the barrier function for the moisturizer, and physiologic lipid mixture can currently be considered as the most ideal moisturizer (Park, 2007a), and it reinforces the lipid barrier function to adjust moisture content of stratum corneum, and intercorneocyte lipid component fulfills this function (Chang *et al.*, 2007). Physiologic lipid mixture is effective in restoring damaged skin barrier, and for moisturizer made of proper composition between ceramide, cholesterol and fatty acid that are physiological lipids of skin, it is absorbed into epidermal cells on epidermis underneath

the barrier layer through skin barrier, forming multi-layer lipid envelope of LB and discharged as intercorneocyte lipid of skin barrier again.

The developmental direction of moisturizer is to find a substance which can normalize the lipid composition and metabolism on stratum corneum based on the biochemical knowledge on stratum corneum that has been accumulated through studies carried out thus far (Park *et al.*, 2001).

Peroxisome proliferator activated receptor (PPAR) has been reported to provide the function to promote keratinocyte differentiation and maintain the homeostasis of skin barrier when it is activated in addition to a lipid metabolism control function, so the development of moisturizer containing components inducing biosynthesis of skin lipids such as intercorneocyte lipid on stratum corneum and PPAR activator is being carried out (Chang & Lee, 2012; Park, 2007a), while for various proteolytic enzymes such as serine protease and aspartic protease involved in the binding force and hydrolysis of capsid protein on stratum corneum, activation is promoted where proper pH conditions and moisture exist, and studies to apply antagonistic conditions to promote or restrain the activation of such proteolytic enzymes are also being carried out (Kim, 2008).

As the non-invasive evaluation of skin barrier function and moisturizing effect became possible, resulting in the development in the studies on skin barrier and the evaluation of moisturizing function of cosmetic products, but the status of skin barrier consisting of epidermal keratinocyte, various lipids and proteins systematically cannot be evaluated properly, sometimes histological or molecular biological evaluations may also be necessary in order to accurately evaluate the barrier function. The human trial is still being most widely used for evaluating the moisturizing function of cosmetic products, both in Korea and abroad (Hong, 2014).

Conclusion

In stratum corneum, corneocyte, considered as the 'bricks' in the 'Bricks and Mortar' model, is connected to specialized lipid and corneodesmosome, the 'mortar' (Kim, 2007), and corneodesmosome which is a protein structure connecting corneocytes, cornified protein envelope and cornified lipid envelope surrounding corneocytes are representative

components that form stratum corneum (Kim *et al.*, 2013). Protein components forming the skin barrier include keratin which accounts for 80–90% of the corneocyte components, profilaggrin/filaggrin involved in the cohesion of keratins after stratum granulosum, precursor protein forming CE (involucrin, envoplakin, desmoplakin, loricrin, keratolinin, SPRRs, cystein-rich protein, cystatin α), cadherin protein forming corneodesmosome, desmoplakin and plakoglobin (Kalinin *et al.*, 2001).

Epidermal keratinocyte goes through the final differentiation process while moving to the upper layer due to the calcium ion concentration gradient (Kim & Jeong, 2012) and proper calcium regulation is very important for maintaining the skin barrier function (Kim, 2010). Also, various cytokines interact with each other on the skin, and dissonance between cytokines may cause pathologic conditions (Ko, 2014).

Skin moisturizing is closely related to the moisture content of the stratum corneum. A decrease in the moisture content of the stratum corneum causes skin dryness as well as abnormality in skin barrier function, so the maintenance of proper moisture in the stratum corneum is an essential condition for preventing moisture loss on intercorneocyte lipid (Kim *et al.*, 2015; Kwon *et al.*, 2013). A moisturizer helps metabolic activity in the stratum corneum to be carried out smoothly (Chang *et al.*, 2007) and affects the skin barrier function with various mechanisms. Significantly, as studies have indicated that it has a positive effect on the differentiation protein of epidermal keratinocyte and lipid synthesizing enzyme (Kim & Jeong, 2012; Park, 2007b), it is expected that a new moisturizer which can restore lamella structure effectively can be useful in treating various skin diseases accompanying xerosis as a adjunctive therapy (Park *et al.*, 2003).

For a correct understanding of the basic concept of skin barrier function and more advanced studies, studies should be carried out to classify various defense functions of permeability barrier and skin and identify the correlation between these functions (Youm, 2013). The ultimate objective of skin barrier research is the future development of cosmetic products including medicines and moisturizers based on the findings (Kim & Jeong, 2012).

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국문초록

표피의 피부장벽

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표피의 각질형성세포가 세포분열과 분화를 거쳐 형성하는 각질층은 피부장벽의 핵심적인 조직이며 이러한 각질층의 형성과정에서 발생하는 불균형과 이상현상은 피부장벽의 기능 이상을 초래한다. 각질층의 기능을 조절하는 인자의 보다 상세한 이해를 위해, 본 논문은 피부장벽을 구성하는 각 요소를 자세히 서술해보았다. “Brick & Mortar” 모델에 따르면, 층판소체, 각질세포막, 지질, 칼슘, 사이토카인, 보습인자 등과 같은 피부장벽의 주된 요소가 자세히 묘사되어 있다. 각질층의 구조와 기능을 설명하는 가장 대표적인 모델은 “Brick & Mortar”로 설명되는 양분 모델이다. 과립층의 각질형성세포 내에는 두 가지 유형의 과립이 있는데 단백질로 채워져 있는 각질유리과립과 지질로 채워진 층판소체가 그것으로, 각질단백막과 각질지질막의 복합체인 각질세포막이 이러한 각질형성세포를 둘러싸고 있다. Filaggrin은 케라틴을 서로 엉겨붙게 하는 접착제의 역할을 하는 동시에 각질세포막을 구성하는 단백질의 결합에도 관여한다. 피부 지질은 장벽 기능으로서 중요한 역할을 하며, 더불어 항균작용 등의 방어기전과 함께 신호 전달 물질로서 피부에 다양한 영향을 미칠 수 있다. 칼슘은 lamellar body (LB)의 구조유지와 세포간극으로의 분비에 관여하며 칼슘 농도구배는 피부장벽의 형성에 결정적인 역할을 한다. 피부장벽이 손상될 때, tumor necrosis factor (TNF), interleukin 1 (IL1), interleukin 6 (IL6)라고 불리는 사이토카인 물질이 분비되는데 장벽의 손상이 장기간 지속적일 때 이러한 사이토카인은 염증이나 표피화과 같은 결과를 야기한다. 과거 보습제의 개념이 피부에 수분공급을 하는 것에 그쳤다면, 최근에는 단순한 수분공급을 넘어 피부장벽기능의 회복에 중점을 두고 있다. 이러한 측면에서 볼때, 생리적 지질 혼합물(physiological lipid mixture)은 현재 가장 이상적인 보습제라 여겨지고 있다.

핵심어: 피부장벽, 층판소체, Filaggrin, 각질 세포막, 보습제

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中文摘要

表皮皮肤屏障

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角质层由表皮角质形成细胞通过细胞分裂和分化而形成, 被认为皮肤屏障的一个核心组织。在角质层形成阶段, 产生的异常或失衡会导致皮肤屏障功能的异常。为了更好地理解角质形成每个阶段的因子的功能, 本文对皮肤屏障的主要组件详细地叙述。从说明砖墙模型 (Brick & Mortar) 开始, 详细地叙述了皮肤屏障的主要组件, 即, 板层小体、角质细胞膜 (cornified cell envelope, CE)、脂质、钙、细胞因子 (cytokine)、保湿剂等。砖墙模型 (Brick & Mortar) 为解释角质层的结构和功能的最典型模型。颗粒层的角质形成细胞内有两种形态的颗粒, 一种是富含蛋白质的透明角质颗粒, 另一种是充满脂的板层小体。角质蛋白细胞膜和角质脂质细胞膜一起被称为角质细胞膜 (CE), 这两种形成角质层的边界。丝聚蛋白 (filaggrin) 作为胶粘剂, 粘合角蛋白, 也参与蛋白结合, 从而形成CE。皮肤的屏障脂质作为屏障功能发挥着重要的作用, 它也作为信号传达物质, 发挥抗菌等防御机制的作用。钙参与板层小体的分泌以及结构的维持, 并且钙梯度在皮肤屏障的形成中也起着重要的作用。当皮肤屏障受损, 会分泌称为细胞因子的物质, 其中称为第一因子的肿瘤坏死因子 (TNF)、白细胞介素 1 (IL1) 以及白细胞介素 6 (IL6) 等细胞因子是非常重要的。然而, 如果皮肤屏障的受损仍然存在, 这些细胞因子会持续上升, 可能会产生炎症或增殖。保湿的概念在过去被理解为补充水分到皮肤, 但最近把注意力都集中在屏障功能的修复, 而不是简单的水分补充。目前为止, 生理性脂质混合物被认为是最理想的保湿剂。

关键词: 皮肤屏障, 板层小体, 丝聚蛋白, 角质细胞膜, 保湿剂

