Review

MOLECULAR STRUCTURE OF THE EPIDERMAL EXTRACELLULAR SPACES

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The skin is a protective cover that participates in the homeostatic regulation of internal fluids by preventing the passage of water, electrolytes and proteins into the external environment. The functional integrity of the skin is maintained through the interaction of the epidermis with the dermis. The epidermis is a tissue arranged in cellular layers. Those epidermal cells in direct opposition to the dermis are the basal cells. The epidermal basal cells are undifferentiated and endowed with mitotic activity. Obeying unknown stimuli, the basal cells undergo maturation following a differentiation pathway that ends with the formation of the stratum corneum. The stratum corneum is an inert film, made of fully differentiated epidermal cells. These cells are anucleated and contain tightly packed fibrous proteins embedded in an amorphous matrix and thick cell membranes. The structure of the epidermis allows a dynamic renewal of stratum corenum throughout life.

The epidermal basal cells are seated on the basal lamina (basement membrane) and are attached to it by an as yet undefined "glue substance." This substance(s) fills the space known as lamina lucida, an electron-lucent space that separates the basal cell membrane from the basal lamina. The basal cells, differentiating keratinocytes and the stratum corneum are attached to each other by another undefined "glue substance." This substance(s) fills the epidermal intercellular spaces (ICS).

The epidermal cells are enclosed in a space limited externally by the stratum corneum and internally by the basal lamina (Fig. 1). Molecules moving in and out of the epidermal compartment must permeate these biological filters. A review of the cutaneous basal lamina was recently published in this Journal,[1] and excellent articles on permeability of the stratum corneum are also reported elsewhere.[2,3] The aim of this article is to suggest a hypothetical "molecular model" for the epidermal intercellular spaces (ICS). This model integrates some known structural aspects of the ICS and recent findings in the field of cell surface biology. Although the biochemical nature of the ICS remains relatively unexplored, recent advances in cell surface biology and epidermal pathophysiology strongly suggest that these spaces are important to maintain a functionally normal skin. An attempt will be made to explore the complex cell adhesion phenomenon and its relevance in epidermal cell pathology. We

Supported by NIH grant 1 R01 AM 21608-01A1.

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0011-9059/79/0700/0434/$00.95

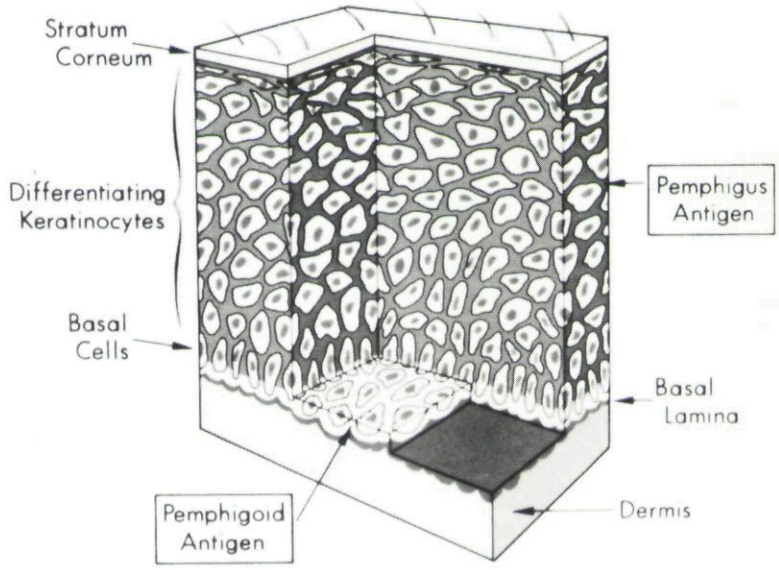


Fig. 1. Structural aspects of human skin.

acknowledge that some of the concepts developed in the course of this review probably are an oversimplification to what may be happening in vivo in the dynamic epidermis.

Cutaneous Basal Lamina

Studies by Briggaman[4] and others[5–8] have shown the cutaneous basal lamina (CBL) to be of epidermal origin. Little is known about the chemical nature of the CBL; however, by analogy to other basement membranes, it is assumed that it has a collagen Type IV chemical composition.[9–11] Two functions are attributable to basement membranes.[12,13] The first function is the provision of mechanical support to overlying epidermal cells. For example, in the basement membrane zone of the chick cornea, a cell to tissue-substrate framework support is formed. This framework may result from macromolecular interactions, presumably ionic in nature, between chondrotin sulfate protein (basal cell surface) and the collagen of the basement membrane (Type IV collagen) and/or that of the primary corneal stroma (Type I collagen).[14] The supramolecular structure thus formed would adhere and confer mechanical support to the overlying epithelia.[15] The second function is the regulation of the molecular movement between compartments separated by this membrane in a fashion perhaps not unlike glomerular filtration in the kidney. Preliminary work on the permeability of the CBL suggests that filtration of molecules across this membrane may be a molecular size-charge phenomenon.[16–18] It is important to note that certain autoantibodies (pemphigus, pemphigoid) as well as complement components, must permeate the CBL in order to reach their cell surface target. Nothing is known about the regulation of the cell locomotion phenomenon across the CBL.

Epidermal Extracellular Spaces

The epidermal and dermal compartments can be further subdivided by the individual cell membrane boundaries into intracellular and extracellular compartments. The latter compartment is also called the intercellular or extracellular space (ICS). The epidermal extracellular compartment is reduced because of the close contact between differentiating epidermal cells as well as between basal cells and the CBL. The space between the basal cell membrane and the CBL is also called the lamina lucida. It is this space which contains the "viscous bond" responsible for the attachment of epidermis to the basement mem-

brane.[19–25] The nature of the "viscous bond" is unknown. Bullous pemphigoid antigen is a normal component of this space.[26] Substances filling this space including pemphigoid antigen are intimately associated with the outer leaflet of the basal cell membrane.[4,14–16,27,28] The cell surface of epidermal cells, melanocytes, Langerhans' cells, and Merkel cells, occupying the epidermal compartment are in close proximity to each other and share a common epidermal ICS which is continuous with the lamina lucida.[16] Morphologically, the epidermal cells are separated from the extracellular space by a trilaminar cell membrane unit.[29] The intermembranous space between adjacent cells are between basal cells and CBL (lamina lucida) is filled with a material which, upon histochemical staining, appears to be rich in carbohydrates. This carbohydrate-rich coat is closely connected to the outer leaflet of the cell membrane[30–34] and constitutes the so-called glycocalyx.[30] At irregular intervals, adjacent cells share common organelles called desmosomes or macula adhaerens.[35] A desmosome is formed by two dense proteinaceous plaques, parallel to the inner leaflet of their respective cell membranes.[36] A bundle of tonofilaments converges from the cytoplasm into the desmosome plaque, looping back into the cytoplasm.[37] Although the fibrilar component of the desmosome unit has been implicated in the formation of the fibrous component of keratin,[38] its relationship to other filamentous proteins such as actin, myosin, tubulin, etc., cannot be discounted.[39] Epidermal cells have been found to contain microtubules[40] and actin and myosin.[41–43] However, their relationship to tonofilaments and epidermal cell membranes remains to be determined. The basal cells show only half a desmosome; their morphology, however, is similar to that of the basic desmosome unit of higher epidermal cells.[28]

The epidermal cell membrane undergoes major changes during differentiation (keratinization),[29,44] and the desmosomes are involved in this process.[45–47] Mitosis and migration of basal cells are accompanied by autophagocytosis of the desmosome unit.[45,48] At the level of the stratum granulosum and stratum corneum, there is a thickening of cell membranes and intercellular spaces and lysis of desmosomes.[29,45] Thus, the intercellular spaces at this level contain, in addition to the degraded desmosomes, enzymatic products from membrane coating granules,[45,47] and constitute, along with keratin,[49] the stratum corneum, the final product of epidermal differentiation.

In 1972, Singer and Nicolson[49] proposed the presently accepted model of cell membrane structure, the fluid mosaic model, based primarily on thermodynamic considerations. Glycoproteins and glycolipids are associated with cell surfaces forming the glycocalyx of cells. The glycocalyx contains a complex set of components such as receptors for hormones, antibodies and lectins, as well as blood group and HLA antigens. The epidermal cell surface contains HLA, blood groups and pemphigus antigen(s) and receptor molecules for lectin and hormones.[51–60]

It has been clearly established that the cell surface—cytoskeletal system interactions play a critical role in many aspects of cell behavior, such as cell recognition, cell adhesion, cell locomotion, cell morphology and embryologic differentiation.[61–74] Any defect in the molecular structure of the cell surface or disruption of the cytoskeletal system (actin, myosin, tubulin, etc.) may trigger pathologic changes in cells, causing them to display abnormalities in their biologic behavior, such as cell adhesion.[71,75,76] Many of these abnormalities are also found in transformed cells.[64,76–81]

In the epidermis, adjacent epidermal cells share desmosome units composed of tonofilaments, attachment plaques, cell membranes and intercellular substances(s). These components are bound together, constituting a firm point of attachment between epidermal cells.[82] The molecular aspects of this phenomenon remain unknown. Cell recognition in the epidermis is shown during the de-

velopment of desmosomes. These studies demonstrate a lack of desmosome formation between unrelated cells such as epithelial cells and pigmentary[83] or Langerhans' cells.[84]

Cell Surface–Cell Adhesion

At the molecular level, the in vivo cell-to-cell or cell-to-substrate adhesion is a complex cell surface biochemical phenomenon. Several hypotheses have been postulated to explain this phenomenon. Tyler and Weiss[85,86] thought that the cell membranes of adjacent cells contained antigen and antibody-like molecules on their cell surface. The formation of immune complexes between the surface of opposite cells would result in intercellular adhesion. Later Roseman[87] postulated the enzyme-substrate theory to explain cell adhesion, in which a cell surface glycosyl transferase enzyme system would catalyze the specific transfer of simple sugars to acceptor-substrates. As a result of this enzymatic activity, the adhesion of cells might occur. Recently, Bell[88] has proposed that cell adhesion may be mediated by formation of reversible bonds between cell surface molecules.

Two types of cell adhesion have been described in vitro[89]: (a) nonspecific adhesion between cells and substrates, such as glass, plastics, collagen-coated slides, etc., and (b) intercellular adhesion which may be specific, if the interacting cells are homologous, or nonspecific, if the cells involved are heterologous. The adhesion phenomenon studied with the use of the tissue culture system thus appears to depend upon a complex, multistep biochemical phenomenon[90–92] in which interactions between serum factors,[93,94] cell surface microexudates,[95–98] Ca++ and Mg++[99,101] and environmental physical factors such as temperature are important.[89,102–104]

Certain purified cells surface antigens ("aggregating factors")[105–112] or cell membrane fractions[113–115] have been shown to promote specific cell adhesion when added to cells of the type from which the factor was extracted. Isolated cell surface glycoproteins involved in cell adhesion have also been described in slime molds,[116] fibroblasts (LETS protein),[98,100,117–122] neural cells,[123–125] etc. Furthermore, cells deficient in these cell surface glycoproteins[75,126,127] show an abnormal, rounded morphology and impaired adhesiveness. Upon reincorporation of the deficient cell surface glycoproteins,[75,76] the cells return to their normal morphology and regain their normal adhesion properties. Cell trans-

Table 1. *Epidermal Cell-To-Cell Adhesion*

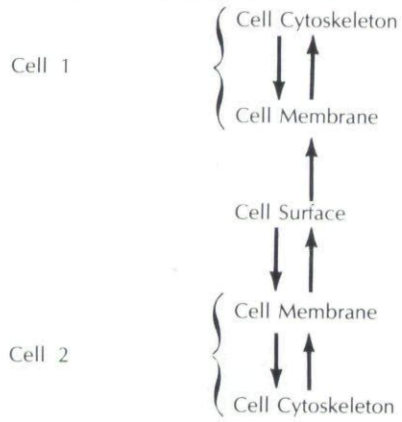
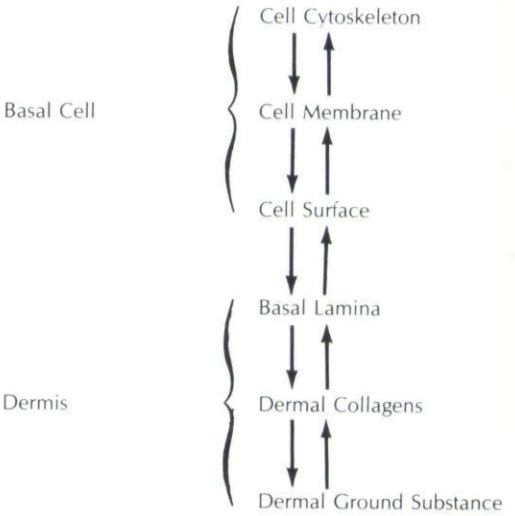


Table 2. *Epidermal-Dermal Adhesion*



formation is associated with a decrease in those cell surface glycoproteins[79,128,129] and with defective cell adhesion.[75–81,91] Immunological studies have provided further information implicating certain cell surface molecules in the cell adhesion phenomenon. Antibodies produced against cell surface antigens are known to induce the following changes when added to antigen-bearing cells: (a) cellular cytotoxicity in the presence of complement,[130,131] (b) induction of antigenic modulation,[132] (c) cell surface aggultination with a corresponding increase in microvilli,[133–135] (d) impaired phagocytosis,[133,134] (e) impairment of inhibition of cell adhesion (aggregation),[76,124,125,135–138] (f) prevention of the restoration of morphology and adhesiveness in transformed cells upon the addition of the deficient cell surface glycoproteins,[75,76] (g) alteration of the normal morphology of cells causing them to become rounded,[76,135] and (h) alteration of embryogenesis.[139]

The types of adhesion phenomena that need to be explored in the skin are: (a) the cell-to-cell adhesion between epidermal cells and (b) the adhesion of basal cells to the dermis. Both are possibly related to a cell surface "glue" produced by the epidermal cells. The nature of this "glue" and its interactions with other unrelated molecular structures (intracellular and extracellular) remain to be studied in the future. Two oversimplified outlines that we follow when studying cell adherence in the skin are shown in Tables 1 and 2. The molecular dissection of each of the components listed and the factors that regulate their interactions probably are important in maintaining the normal epidermal cell adhesion in vivo. It is predicted that any disturbance in each of these interacting systems will be manifested in a clinical disease. Thus, "dissolution" of the cell surface "glue" that normally attaches differentiating keratinocytes is found in diseases such as pemphigus vulgaris, Hailey and Hailey and certain forms of toxic epidermal necrolysis. "Dissolution" of the basal cell surface "glue" that attached these cells to the basal lamina is observed in the skin of patients with bullous pemphigoid, and epidermolysis bullosa letalis. These are a few of the dermatoses that maybe considered "experiments" of nature and from which we may learn important aspects of epidermal cell biology.

Dynamic Concept of the Epidermal Extracellular Spaces

In addition to cell membrane components, the epidermal extracellular spaces may contain molecules of epidermal and extraepidermal origin (external environment, serum, dermis) which have permeated the CBL or stratum corneum. The molecular structure of the epidermal ICS and the way it may regulate the movement of the extracellular molecules[140,141] remains to be explored. The ICS maybe conceived as a dynamic "biological molecular mesh" formed by interacting cell surface molecules of cells sharing the epidermal compartment. This dynamic state would depend on the integrity of the epidermal and nonepidermal cell membranes, the permeability of the stratum corneum and the basal lamina, and perhaps the degree of differentiation, and the metabolic state of the cells enclosed in this compartment. The importance of the permeability of the CBL and those factors that may control or regulate its filtration properties remains unknown. The CBL may play a critical role in keeping epidermal antigens away from pathogenic cytotoxic antibodies as postulated by Terasaki and Chamberlain.[142] These authors found that autologous serum was cytotoxic to epidermal cells in vitro. Thus any disturbance in the CBL permeability may cause accumulation of fluid, penetration of cytotoxic autoantibodies and chemotatic cell migration into the altered epidermal extracellular compartment.

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