INDIAN JOURNAL OF DERMATOLOGY, VENEREOLOGY AND LEPROLOGY VOL. 42, NO. 4, 1976

A BIOLOGIST LOOKS AT SKIN STRUCTURE

H. PINKUS

Anatomical investigations of the skin have been carried on for many decades. Interpretation of structural details in scientific biological terms, on the other hand, goes back only 25–30 years, and its modern progress was greatly stimulated by S. Rothman's pioneer text on biology and biochemistry of skin published in 1954. I had the good fortune of being exposed to H. Spemann's lectures on experimental embryology and his concepts of tissue and organ interactions early in my medical studies and later became interested in explanation of tissues and studied with some success the behaviour of living human epidermal cells in tissue culture from 1930–1933.

I'd like to discuss today some of the biological concepts which we need in order to understand normal skin structure and its pathological alterations as seen in our fixed and stained sections. The skin has at least four basic functions: it is the all important barrier between hostile surroundings and our bodies and maintains homeostasis; it is the organ of perception for pain, temperature and touch stimuli; it has a multitude of metabolic activities, perhaps more diverse than those of the liver; and it reacts in a variety of ways to injury of all kinds, mechanical, chemical and infectious, and is indeed a principal organ in immunological processes. The basis for investigation of all these functions must be through knowledge of skin structure and skin biology.

We must realize from the beginning that to speak of "The skin" is an abstraction. There are many different types of skin: thickness and toughness of epidermis and dermis vary tremendously between eyelid and sole of the foot; number and size of hair follicles, eccrine, apocrine and sebaceous glands vary similarly from scalp to face to trunk and to various portions of the extremities. Biologic and functional aspects show corresponding variations.

Many normal and pathological functions of the skin are concentrated in the relatively thin superficial portions comprising epidermis and papillary dermis. Epidermal thickness on most of the body surface is less than 0.2 mm, and it consists entirely of epithelial cells without any supporting extracellular fibres. Yet it forms a tough and resilient external coat and an almost complete barrier to exchange of water and chemicals. It also undergoes continual renewal of its elements, keratinized cells being exfoliated daily and replaced by new cells from below. I first became more intensely interested in growth and turnover of the epidermis in 1950 because there were published reports contending that mitotic activity in the epidermis was too low to explain this constant turnover, and that lymhocytes moved into the epidermis from

---

This was delivered as an oration at the 4th Annual Conference of Indian Association of Dermatologists, Venereologists and Leprologists, held at New Delhi in February 1976.

Professor of Dermatology, Wayne State University, Detroit. Michigan (USA).

below, transformed themselves into "clear cells" and then into epidermal cells. Basing my views on my earlier experiments with tissue culture which had shown the epidermis to consist of motile cells quite capable of mitotic division. I set out to disprove these improvable conclusions by quantitative analysis. I used pressure-sensitive adhesive tape to strip off the stratum corneum with the intent of counting the number of horny layers and the number of keratinized cells. To my elated surprise I discovered that simple mechanical removal of the "dead" layers provoked a tremendous burst of activity in the basal layer. As many as 4–5% of all cells were undergoing mitotic division in specimens taken 48 hours after the injury, enough to replace all epidermal cells within 24–30 hours. Furthermore, a simple diagram illustrating the relative size, shape and number of the various cell types made it obvious that nature has constructed the epidermis with great economy in order to achieve a perfect functional result. All mitotic activity is restricted to the basal layer, which is the matrix or germinal layer of the epidermis. When a daughter cell moves up and loses contact with dermis, it becomes a 'prickle' cell and sets out on a path of maturation involving intricate synthetic processes, which ends with the conversion to a non-nucleated corneocyte in the horny layer. During this slow conversion, which normally requires about 4 weeks, the shape of the cell changes from a slender column through a bulky multifaceted body to a flat pancake-like flake. The transverse diameter increases from 6–10 microns to 30–45 microns. Assuming that both surfaces are flat, we find the same area that is occupied by approximately 100 basal cells at the bottom needs only 4 corneocytes in the top layer. Many investigators have confirmed and extended these results by sophisticated methods, and it is quite clear now that a low mitotic index of 1:1000, or even 1:10,000 is quite sufficient to explain the exfoliation of one horny layer per day. The stratum corneum may be 15–30 layers thick on the average body surface, against 6–10 layers of living cells up to the stratum granulosum, yet it contains only about 15% of the total number of cells.

When we refer to keratinized cells (corneocytes) as dead, we must realize that the non-nucleated erythrocytes also are dead in a biological sense, yet we consider this end-product of the bone marrow as the vital goal of all biological activity in the hematopoietic system. Similarly, the horny flake is the result of complicated synthetic processes going on in all layers of the epidermis and being unravelled only now by the work of numerous investigators. The precursor proteins in tonofilaments and keratohyalin granules finally become consolidated into the intricate stucture of insoluble keratin to accomplish the barrier function of the skin without which mammalian life in general and human life in particular would be impossible. We therefore, call the entire clan of epidermal cells properly keratinocytes in order to express their biological destination. Bullough and his associates called attention to yet another function of the stratum corneum. It contains chalones, mitosis-inhibiting substances, which act as a brake on epidermal turnover. Removal of corneocytes by stripping or pathological processes releases the brake and leads to greatly increased mitotic activity and rapid epidermal turnover with corresponding decrease in the life span of the keratinocytes. Contrariwise suppression of basal cell mitosis, which can be accomplished easily by *weat irradiation*, e.g. thorium leads to accumulation of granular and horny cells, prolonged lifespan of keratinocytes and a shift in proportion of young and old cells. How these

experimental data can be applied to the interpretation of skin diseases has been explained in previous publications on psoriasiform and lichenoid tissue reactions.

When we call the cells between basal and granular layers prickle cells, we actually use a misnomer which, however is so ingrained that it would be foolish to try to change it, especially since terms like acanthosis, acantholysis and acanthoma are based on it. The keratinocyte at this stage does not resemble a ball beset with prickles. If a cell becomes isolated in pathological processes or in tissue culture, it rounds off into a smooth surfaced sphere, and only electron microscopy has shown recently that its surface may carry microvilli. What early anatomists interpreted as prickles are cross sections of inter-cellular bridges which form only when two or more keratinocytes are in close contact. The details of these inter - cellular connections have been worked out by electron microscopists who have pointed out that those conditions under which bridges and tonofibrils are best visible to the light microscopist, that is in acanthotic epidermis of chronic inflammatory conditions or condyloma acuminatum, are in fact pathological. Normal keratinocytes in well fixed specimens are closely apposed with interlocking folded cell membranes, which in numerous places are connected by intricately structured desmosomes (gap junctions), to which the tonofilaments stream from the cytoplasm. When intercellular spaces widen under conditions of mild edema, then the desmosome-carrying stretches of cell membrane remain tied together and portions of the cytoplasm are drawn out to form the two halves of a bridge. The question arises, how permanent are desmosomes? Their intricate layered structure suggests great stability. It is known that they split in acantholytic disease with dire consequences for the viability of the epidermis. Inasmuch as there is constant, if slow movement of cells against each other even in normal epidermis, and as we know that leukocytes can move fairly freely between the keratinocytes, we must postulate that desmosomes are not permanent. Nothing however, was known concerning the time it might take to dissolve or re-form desmosomes. In order to obtain data, Mishima and I used the tape stripping procedure in which movement of cells against each other obviously is accelerated even before mitotic activity sets in. We found that 2-4 hours after the injury, inter-cellular connections edema is obvious under the electron microscope, and inter-cellular connections are broken in many places. Contrary, however, to the conditions in pemphigus, desmosomes do not split at the cementing line. Breaks occur in the cell membrane next to desmosomes and intact desmosomes remain attached to one of the two cells. Later they are taken into the cytoplasm and degraded. The cell surfaces develop numerous microvilli and after 18-24 hours, it seemed that microvilli establish new connections between neighbouring cells, and new desmosomes develop. The process thus appears to be a matter of hours rather than of minutes or days.

Literature concerning the seat of the barrier in epidermis is large. It seems that the entire stratum corneum is concerned, perhaps by acting as a spongy filter that permits only slow progress of solutes in water or lipid. There is, however, evidence that the barrier for water itself is more definitely situated just above the granular layer. When the stratum corneum is stripped away completely water evaporates from the skin as from an open vessel. The water barrier is re-established after 24-48 hours, when a keratotic surface layer is formed. Normal skin exhibits a glassy lucent layer above the stratum granulosum. Although some text-books state

that this stratum lucidum is present only on palms and soles, that statement should be modified to say that a stratum lucidum is easily demonstrable in these areas by routine methods, but a thin layer is visible by special staining (e. g. congo red) in many tissue sections, and the biological processes which convert a granular cell into a fully keratinized cell do, of course, go on everywhere and must pass through that functional arrangement of lipids and proteins which in thick epidermis appears as the peculiarly glassy stratum lucidum. Finally, let us look at the other barrier that seems to exist at the lower epidermal surface, the often quoted basement membrane, or in a more general term, the dermo-epidermal junction. Old controversy concerning the existence of a structural basement membrane has been resolved by modern investigations, and as so often in biology the informed answer is both yes and no. There is no thick hyaline membrane. Rather there is a fairly broad basement zone, in which pedicles of basal cells interlock with reticulum and modified elastic fibrils as if the bristles of a brush were pushed into the pile of an Indian carpet. Furthermore, the basement zone is impregnated with neutral mucopolysaccharides, which appear as a continuous band in PAS stained sections. This is the structure revealed by light microscopy. Electron microscopy shows the basal cell membrane paralleled by a basal lamina with translucent and dense layers. Hemidesmosomes and anchoring fibrils provide further support. The epidermo-dermal junction thus has a complicated structure insuring firm cohesion that is disrupted only in certain pathological circumstances or by experimental procedures.

The basement zone, however, is not a complete biological barrier. It may be termed a filter or a sluice. Water and water soluble substances quickly penetrate it as can be shown by injecting certain dyes, radioactive aminoacids or thymidine, and even small particles like ferritin into the dermis. Larger molecules, such as globulins and complement, may adhere to the basement membrane and can be demonstrated by immunofluorescent methods in various diseases. Living cells, especially leukocytes, easily penetrate into the epidermis from the dermis. Basement membrane which in part is formed by the epidermis, in part by the dermis, in fact is evidence of ectodermal-mesodermal interaction and co-operation. It begins to be formed in mid-fetal life, and it disappears again under conditions of invasive neoplasia. It is wrong to say that cancer cells "break through" or "destroy" the basement membrane. They are just incapable of forming or inducing it, and therefore, I have tried to give a few glimpses into biological interpretation of skin structure with the intent of providing better insight into normal and pathological histomechanisms in the skin and with the hope that such insight may lead to advanced principles of therapy :

REFERENCE

1. Pinkus H and Mehregan AH : A Guide to Dermatohistopathology, 2nd edition. Appleton-Century-Crofts (Prentice-Hall) New York, 1976.