

SHORT REPORTS

Basal Keratinocyte Subsets: Ultrastructural and Morphometric Features

MAURIZIO ZAMPETTI, ANDREA FATTOROSI, TERESA GRIECO and STEFANO CALVIERI

Istituto di Dermatologia, Università "La Sapienza", Rome, Italy

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Basal keratinocytes reportedly comprise two ultrastructurally recognizable populations, usually referred to as serrated basal keratinocytes (SBK) and non-serrated basal keratinocytes (NSBK). The former are responsible for dermal-epidermal anchoring, whereas the latter serve in epidermal turnover (stem cells). The size and shape of these cells were investigated by electron microscopy and computer-assisted morphometric analysis. The results showed that perimeters of both the nucleus and cytoplasm of NSBK were consistently smaller than those of SBK. The shape factor (contour index) was lower in NSBK, indicating a more regular membrane surface. Present quantitative results substantiate previous subjective reports on the ultrastructural differences between NSBK and SBK and provide additional evidence that the basal cell layer in human epidermis is actually made up of at least two types of morphologically distinct keratinocytes. (Accepted May 19, 1988.)

S. Calvieri, Clinica Dermatologica, Università di Roma, Viale dell'Università, 00185 Roma, Italy.

There is now convincing evidence that two distinct basal keratinocyte subsets exist in human palmar epidermis (1-3). These two cell types have been distinguished on the basis of their location and morphology, and are referred to as serrated basal keratinocytes (SBK) and non-serrated basal keratinocytes (NSBK) (1, 2). SBK have the characteristics of typical basal keratinocytes and are responsible for dermal-epidermal anchoring, whereas NSBK are reportedly responsible for epidermal cell turnover (4-6).

In spite of this ample body of knowledge, we are not aware of any systematic morphometric study of SBK and NSBK in human skin. The aim of the present paper was therefore to investigate by electron microscopy normal human palmar epidermis in order to identify and further characterize these keratinocyte subsets, and to quantify their typical morphological features by computer-based image analysis technique.

MATERIALS AND METHODS

Microscopy

Biopsy specimens taken from 4 healthy volunteers' palmar skin served for this study. Samples were processed according to standard procedures (7). Briefly, specimens were fixed in buffered formalin-glutaraldehyde, post-fixed in buffered osmium tetroxide and stained with methylene uranyl acetate and lead citrate.

Morphometry

Data were acquired by scanning 50 cells, either NSBK or SBK, from electron microscope photographs (final magnification $\times 4500$) using a light pen and a graphic tablet. Evaluation of data was performed off-line using an Apple IIe microcomputer. The measurements were performed by tracing the contours of cytoplasmic and nuclear membranes as they appeared on micrographs. Only sharply and completely outlined keratinocyte profiles were traced, and the digitized information was used to calculate the sizes of cells and nuclei. Cytoplasmic and nuclear area as well as cell perimeter and maximum diameter were measured. Nuclear/cytoplasmic (N/C) ratios for area and perimeter were calculated from mean values.

In the present work, data are expressed as computer-based arbitrary units. As a shape-measuring parameter, the cellular contour index (CI), was calculated from mean values. CI is a size-independent measurement of the shape of a profile. It is calculated using the following formula: $CI = \text{Cytoplasmic perimeter} / \text{cytoplasmic area}$. A circle has a CI of 3.54.

Increasing irregularity of a profile produces higher values (8). Thus, in the present paper, CI quantifies the degree of convolution of the cell surface. Statistical evaluation of samples was performed with the Mann-Whitney test.

RESULTS

Electron microscopy

SBK were the only keratinocyte subset found in epidermal crests corresponding to the less deep stratum corneum sulci. They were cylindrical, closely packed and had the well-known ultrastructural morphology of typical basal keratinocytes, with few mitochondria and ribosomes, evident melanosomes and a cytoplasm particularly rich in tonofilaments, arranged either in tonofibrils or in thick bundles (Fig. 1a). Additional features included long, cytoplasmic projections most evident on the dermal border, and several desmosomes and hemidesmosomes. The oval nucleus contained a small nucleolus and finely dispersed granular chromatin. Dermal anchoring fibrils were well represented. NSBK were found exclusively at the bases of one or more of the deeper epidermal crests corresponding to each stratum corneum ridge. These cells presented a cuboid or oval shape, abundant cytoplasm rich in mitochondria, ribosomes and rough endoplasmic reticulum. These organelles were frequently arranged around the nucleus (Fig. 1a). Tonofilaments were rarely seen. The cytoplasmic membrane presented variable amounts of villi-like projections and the dermal border showed short thick protrusions. Only a few desmosomes and hemidesmosomes were observed, and particularly scarce were the dermal anchoring fibrils. Nuclei were centrally located and contained finely granular chromatin irregularly clumped at the periphery (Fig. 1b).

Morphometry

Data from morphometric analysis are summarized in Table I. The cytoplasmic and nuclear area, and Nuclear/Cytoplasmic area ratio of SBK were greater, on the average, than those of NSBK but did not reach statistical significance. Cytoplasmic and nuclear perimeter, and maximum diameter of SBK were significantly greater ($p < 0.05$) than those of NSBK. The CI showed that SBK had a much more irregular outline than NSBK ($p < 0.05$).

DISCUSSION

NSBK and SBK are easily distinguishable on the basis of their peculiar ultrastructural features (1-3). The present data provide quantitative analysis of several sizes of these cells, and

Table I. *Morphometric parameters of SBK and NSBK (mean \pm SD, arbitrary units)*

Variable	SBK	NSBK
Nuclear area	23.75 \pm 5.10	18.51 \pm 4.20
Nuclear perimeter	18.58 \pm 4.30	12.45 \pm 2.86*
Cytoplasmic area	80.50 \pm 11.41	70.32 \pm 10.81
Cytoplasmic perimeter	83.93 \pm 13.27	55.48 \pm 8.84*
Max. diameter	16.99 \pm 2.01	11.239 \pm 1.88
Nuclear/Cytoplasmic ratio	0.295 \pm 0.40	0.263 \pm 0.10
Nuclear/Cytoplasmic perimeter ratio	0.221 \pm 0.32	0.224 \pm 0.32
Cytoplasmic Contour Index	9.35 \pm 0.88	6.62 \pm 0.91*

* $p < 0.05$.

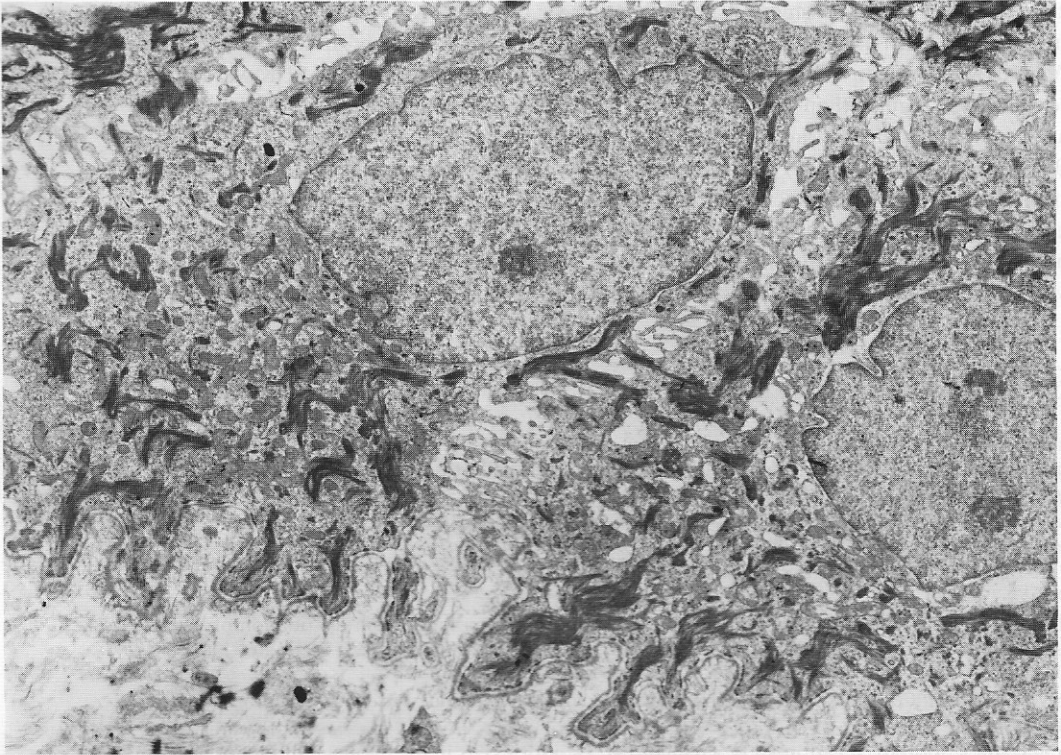


Fig. 1 a. Representative view of typical serrated basal keratinocytes (SBK). Note the numerous melanosomes and thick bundles of tonofilaments. Cytoplasmic membrane has a very irregular outline, particularly evident at the dermal-epidermal junction, where elongated projections are visible ($\times 10\,000$).

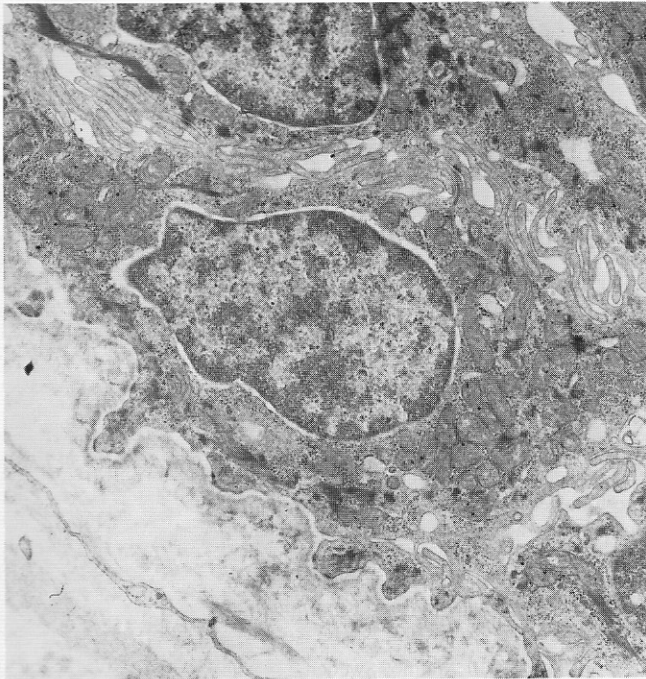


Fig. 1 b. Representative view of typical non-serrated basal keratinocytes (NSBK). Note the numerous mitochondria, ribosomes, and cisterns of rough ER. Cytoplasmic membrane exhibits short villi-like projections. The dermal surface, however, is regularly shaped ($\times 10\,000$).

confirm the subjective assessment that SBK are larger than NSBK in both nuclear and cytoplasmic perimeters. The most relevant morphometric differential feature, however, is the CI. In keeping with subjective judgement, the cytoplasmic membrane profile of SBK was found to be much less regular than that of NSBK. This highly indented profile is fully compatible with the dermal-epidermal anchoring role reportedly played by SBK. On the other hand, NSBK have smoother borders and few desmosomes, strongly suggesting that these cells do not fulfil any fundamental function in dermal-epidermal anchoring mechanisms (1-6).

In conclusion, the present results indicate that NSBK and SBK are readily detectable in human palmar skin by electron microscopy, and that quantitative techniques can be useful in providing an objective means for the assessment of size and shape differences between these two keratinocyte subpopulations.

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Implantation of Orthopaedic Devices in Patients with Metal Allergy

ÅKE CARLSSON and HALVOR MÖLLER

Departments of Orthopaedics and Dermatology, Lund University, Malmö, Sweden

Å. Carlsson, H. Möller. Implantation of orthopaedic devices in patients with metal allergy. *Acta Derm Venereol* (Stockh) 1989; 69: 62-66.

Patients with a contact allergy to chromium, cobalt and/or nickel, patch test verified before implantation of a metallic orthopaedic device, were followed up years later by clinical and radiographic examination as well as with epicutaneous and intracutaneous tests. Eighteen patients had been exposed to an orthopaedic implant for several years (mean 6.3 years) containing a metal to which they were allergic. None had suffered any dermatologic or orthopaedic complications attributable to the contact allergy. *Key words: Contact allergy; Chromium; Cobalt; Nickel; Bio-implantation.* (Accepted June 9, 1988.)

H. Möller, Department of Dermatology, General Hospital, S-214 01 Malmö, Sweden.