CHANGES IN ADHESIVE INTERACTIONS BETWEEN KERATINOCYTES DURING SKIN REGENERATION IN RATS

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KEY WORDS: skin; adhesion; keratinocytes; epidermis; wound healing

Adhesive interaction between keratinocytes determines the integrity of the epithelial cover and its barrier function.

Information on changes in these interactions when the skin is injured, and also during its regeneration, is important in order to understand both the mechanisms of resistance of the skin and mechanisms of repair processes. Unfortunately, the available data are extremely scanty [2].

The aim of this investigation was to try to fill this gap by studying changes in adhesive interactions between keratinocytes during regeneration of full-thickness wounds in rats.

EXPERIMENTAL METHOD

Noninbred male rats weighing 200-250 g were kept in single cages. Before injury the animals were anesthetized by intraperitoneal injection of pentobarbital (40 mg/kg body weight). The hair was removed in the interscapular region from an area of 24 cm^2. In the center of this area, the skin was tattooed in the form of a circle 17 mm in diameter, using a stencil. A full-thickness, circular piece of skin 16 mm in diameter was excised within this circle. The parameters were recorded in areas covered with epithelium 21, 30, and 60 days after injury, in regions of scar tissue, of the peripheral epithelium (the region between the tattooed surface and the wound edge), and neighboring undamaged areas of skin, located not less than 1-2 cm from the wound. The force of adhesion (F, g/cm^2) between the epidermocytes was measured by means of an "Epitest" adhesimeter ("Biotekh-Élektron," Moscow), detaching a probe, glued to the epidermis with cyanoacrylate glue, at the rate of 100 mm/min [1]. The procedure was repeated six times in the same place, with layer by layer penetration into the depth of the epidermis. The results obtained on removal of the first layer were disregarded.

To assess the barrier function of the epidermis the resistance of an area of skin to an alternating current (1 kHz) was determined by means of Ag–AgCl electrodes of membrane type on an "Epilar" apparatus ("Biotekh-Élektron," Moscow). The number of animals in the group was at least five. Statistical analysis of the data was carried out by parametric and nonparametric tests of significance. All the results were subjected to statistical analysis.

EXPERIMENTAL RESULTS

Observation on the kinetics of changes in resistance of the wound surface during healing revealed that resistance of the wound in the initial period was 10 times less than the resistance of the neighboring intact area of skin (resistance 0.31 ± 0.01 and 3.10 ± 0.80 kΩ respectively, $p < 0.01$). The value of this parameter increased during the subsequent period and on the 21st day it did not differ from the control values, namely 6.4 ± 0.9 and 6.1 ± 0.9 kΩ

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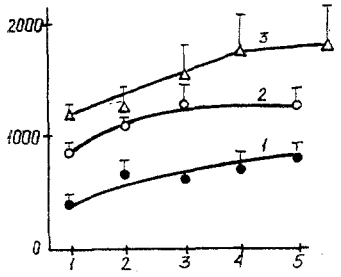

Fig. 1

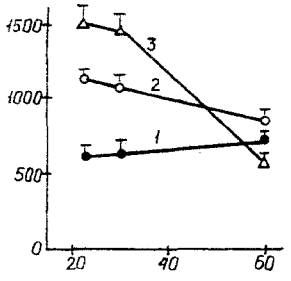

Fig. 2

Fig. 1. Dependence of force of adhesion between keratinocytes on their depth in epidermis, measured in regions of: 1) intact skin, 2) peripheral epithelium, 3) scar on 21st day after infliction of circulation full-thickness wounds to rats' skin.

Fig. 2. Dependence of force of adhesion between keratinocytes on time in regions of: 1) intact skin, 2) peripheral epithelium, 3) scar during healing of full-thickness circular skin wounds in rats.

respectively ($p > 0.1$). Since resistance is determined chiefly by the epidermis [4], the results are evidence that its barrier function was restored after 21 days.

The results of measurements of the distribution of the force of adhesion by depth in the epidermis after 21 days are given in Fig. 1. A relationship similar to that which we found previously [1] was clearly apparent in the intact region, consisting of reduction of the force of adhesion between cells in the direction toward the stratum corneum. The same dependence was discovered in the region of the scar and in the peripheral epithelium. However, absolute values of the force of adhesion in these two regions were several times greater than the control values, and the differences were greatest in the region of the scar. Dependence of the force of adhesion, averaged for a given region, on time of observation, measured in areas of skin of all three types, are given in Fig. 2 [$F = (\Sigma_{i=2}^{6} F_i)/5$].

The value of the force of adhesion in the epidermis of the scar and the peripheral region slowly decreased to reach the control values, but only after 60 days had elapsed.

The data indicate that after complete epithelization, which was achieved after a comparatively short period of time (under 21 days) the process of maturation continued in the outwardly unchanged epidermis, in which it was expressed in particular as a change in adhesive interactions between keratinocytes.

A similar process of maturation of the rat epidermis was observed by the writers previously during the first month of postnatal development [3]. The results showed that absence of desquamation, characteristic of the neonatal epidermis, is accompanied by high values of the force of adhesion, whereas an increase in the rate of desquamation and reduction of the number of cell layers correspond to a marked reduction of the force of adhesion.

Comparison of these data suggests that the rate of desquamation in the newly formed epidermis of the scar and peripheral zone is reduced, or desquamation is absent, during the first 1-2 months after injury.

The observed increase in adhesion probably reflects the adaptive reaction of the skin to injury, with the aim of reducing loss of cells and ensuring the most rapid recovery of the epidermis and its barrier function. The results of our observations indicate that the barrier function, the most important for maintaining viability of the individual, is actually the first function to recover, and only later is it followed by normalization of adhesive interactions between the cells. The duration of maturation of the epidermis, in the sense noted above, exceeds the duration of the period of epithelization of the wound surface.

A similar result was found previously when recovery of the connective-tissue matrix was monitored – the duration of its maturation, expressed in particular as increased breaking strength of the scar tissue, which is several times greater than the duration of formation of the scar itself at the site of the wound [5].

Thus after closure of a wound there follows a period of prolonged maturation not only of connective tissue, but also of epithelial tissue. Is there any difference between the reaction of maturing tissues of altered skin to the additional damaging action (drugs, physical, chemical factors, etc.) and the reaction of damaged tissues? The answer

to this question may be of great practical importance, especially for groups of people whose skin is subjected to unfavorable external environmental factors during life and at work.

Thus as a result of the investigations a reversible increase in the force of adhesion was found between the keratinocytes both in the epidermis covering the scar formed at the site of a full-thickness wound and in the peripheral epidermis. This effect is of adaptive importance, for it can minimize cell losses and ensure rapid restoration of the epithelial cover and its barrier function.

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REPARATIVE CHANGES IN THE SENSOMOTOR CORTEX OF THE OFFSPRING AFTER MODERATE PRENATAL EXPOSURE TO ALCOHOL

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KEY WORDS: moderate prenatal exposure to alcohol; offspring; sensomotor cortex; reparative changes

Recently ever-increasing attention has been paid to the study of the effect of moderate prenatal exposure to alcohol on the development of the fetus and offspring. However, brain morphology has received little study in these cases. Leptomeningeal neuroglial heterotonia has been discovered in the frontal pole and the middle temporal gyrus, a decrease in density of the neurons and gliosis in the superficial layers of the frontal cortex, and reduction and dysplasia of the lateral geniculate body in the 6-month-old offspring of a monkey consuming 2.5 g/kg of alcohol once a week since the 40th day of pregnancy [9]. In the offspring of BALB/c mice with moderate prenatal exposure to alcohol the area of the corpus callosum and of the anterior commissure was reduced [10]. Delayed development and dystrophic changes of the cortical neurons were observed in the offspring of rats receiving alcohol in a dose of 2 g/kg body weight during pregnancy, at the light-optical level [8], together with changes in the ultrastructure of neurons [4], interneuronal connections [5], and capillaries [6] in the sensomotor cortex, lasting until the period of puberty.

In this publication attention is drawn to reparative changes in the sensomotor cortex of the offspring of rats with moderate prenatal exposure to alcohol.

EXPERIMENTAL METHOD

A model of alcoholic intoxication of pregnant females was developed at the Institute of Pharmacology, Academy of Medical Sciences of the USSR, where the experimental offspring were subjected to a preliminary physiological investigation. From the 1st to the 20th days of pregnancy, the mother rats received 20% alcohol

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