The Peculiarities of Thermal Burn Histological Skin Structure After Application of Far Eastern Natural Zeolite Nanoparticles

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Abstract: *At present working out biologically active wound coverings, where it is possible to include the sorbents created on the basis of zeolites, is very perspective direction in developing new types of bandaging material at burns. The peculiarity of zeolites at burns, with its ability to absorb toxic substances, is their participation in regulation of electrolitic homeostasis, and also their antioxidant properties. In this paper the results of research of patients' histological skin structure with II and IIIa degree burns after application of zeolites from the Far Eastern deposits are described.*

Keywords: *zeolites, nanoparticles, skin, burn, antioxidant, nanotoxicology.*

INTRODUCTION

Burns were always and still are one of the most widely spread types of traumatism. Skin plays an important role in protecting the body in the environment from different factors.In literature there is much evidence that physiological and morphological changes of the skin occur even when there are I degree burns (11). Among the substances which are studied in medicine thoroughly there is an interesting group - zeolites (5-7, 9, 12). In literature one can see the data about using zeolites for treating the burns and overheating(1-4).

The aim of our research was to determine the histological skin structure after II and IIIa degree burns and during its correction by using nanoparticles of zeolite from the Russian Far Eastern deposits Vangin, Kulikov and Lyutog.

MATERIALS AND METHODS

The experiment took place in Primorskiy Burning Center (Vladivostok) with the volunteers who had II and IIIa degree burns. All the patients were informed about the experiment and they agreed in a written form to use the applications with zeolites and biopsy.

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All manipulations with people were carried out according to the Declaration of World Medical Association (Helsinki). Before the beginning of the experiment all the volunteers at the age from 25 to 40 were divided into four groups, seven people each.

"Control" - the patients who had applications with "Levomekol" ointment, "V" - volunteers who had powdered zeolite applications on their burnt skin from Vangin deposit (Amurskiy region), "K" - volunteers who had powdered zeolite applications on their burnt parts of skin from Kulikov deposit (Amurskiy region), "L" - volunteers who had powdered zeolite applications on their burnt parts of skin from Lyutog deposit (Sakhalin Island). In order to get the aim zeolite was crushed (the crusher VKMD6) and further it was ground with ultrasound (homogenizer Bandelin Sonopulse 3400). As a result, the size of zeolite particles was about 500 nm. The concentration of zeolite in tuffs of all three deposits was approximately 60-70%. The applications of zeolites (weighing up to 1 g) on the burnt parts of skin were carried out during one week and once a day. After the experiment the patients were exposed to skin biopsy. For histological examination the material was fixed in 10% neutral formalin solution, the filling was in paraffin. Paraffin sections were colored with Boehmer hematoxylin and eosin, PAS-reaction on Mac-Manus, orsein on Uns-Tänzer; semi-thin sections were colored with methylene blue and osmium-zinc iodide on Akert, Sandri. For morphometry we used the microscope Zeiss Axio Image A1 and the program "VideoTesT-Morphology - 5.1. We studied the perimeter, area, length, width of cell and nuclear. Statistical data processing was carried out with the program Statistica 6.0.

RESULTS AND DISCUSSION

In the control group in biopsy material we observed vascular congestion, increased exudation, and as a result, swelling of the dermis, which is the first phase of healing. In the granulation tissue comparing with the normal condition, a large number of neutrophils was observed (8, 11). In group "B" we observed the effective development of granulation tissue. Fibroblasts formed layers and had large sizes. Morphometric parameters of tissue basophils in group "B" were much higher comparing with the control group where patients had standard treatment: the perimeter of the cells increased by 18.2%, the area by 44% and the length by 25% (Table 1).

Table 1. Morphometric data of skin tissue basophils being burnt and treated with zeolite applications

	Groups			
Data	Control	V	K	L
Perimeter, mkm	33,32±1,42*	39,38±1,89*	33,95±1,29**	37,88±1,82**
Area, mkm^2	56,24±2,02**	80,98±4,12*	57,22±1,96*	78,81±4,03*
Length, mkm	14,11±0,53**	17,63±0,95*	14,98±0,55**	16,55±0,79**
Width, mkm	5,76±0,34*	5,68±0,32	5,85±0,33*	5,71±0,28*

* - validity p>0,05; ** - validity p>0,001.

In group V also in comparison with control group morphometric parameters of nuclear and keratinocyte cells (the area by 15%, length by 16%), Merkel cells (the area by 14%, the area of the nuclear by 13%) and Langerhans cells (the area by 21%, the length by 19%) increased.

In the material of group "B" also in comparison with the control group, there was decreasing the height of the skin papillae, loosening of the membrane basement, the percentage of cells vacuolization and thickened collagen and elastic fibers focal accumulations. These data are tally with the data in literature (1, 2).

In groups "K" and "L" in the skin the increase of most indices was observed in comparison with the results of the control group where "Levomokol" was used. The percentage of vacuolization in the skin decreased comparing with the control (by 10% and 22% in the basal, by 16% and 26% in the thorny and in 1,6 and 3 times in the granular layer for groups "K" and "L" respectively) . Morphometric indices of Langerhans and Merkel cells, keratinocytes and tissue basophils in the group "L" were valid to the indices of group "B" (Table 2), whereas in group "K" in comparison with the control group (to 10%)the increase of these indices was slight and not valid statistically .

The quantity of elastic and collagen fibers in group "B", "K" and "L" in comparison with the group "Control" significantly reduced, they became thinner and more streamlined.

Table 2. Some morphometric data of Langerhans and Merkel cells of burnt and treated with zeolite applications skin

	Nucleus				Cell			
Data	Control	V	K	L	Control	V	K	L
Langerhans cell								
Perimeter, mkm	40,22±1,51*	42,66±1,53**	40,05±2,16*	42,71±1,39	53,73±1,32	59,95±1,85**	59,21±1,52**	58,34±2,13
Area, mkm^2	47,42±3,57*	48,37±2,86**	48,18±2,58**	50,76±2,78**	130,14±5,54**	157,47±3,48**	137,99±4,25**	152,21±4,63**
Length, mkm	11,42±1,42	13,51±0,73*	11,75±0,67*	13,44±0,58*	15,92±0,62**	18,95±1,61*	17,37±0,47*	18,87±1,57**
Width, mkm	5,81±0,45*	5,92±0,37*	6,02±0,34**	5,96±0,39**	14,89±1,53*	16,58±0,73**	15,35±0,69**	16,32±1,24
Mercels cell								
Perimeter, mkm	45,57±2,22*	46,12±2,01*	46,35±3,65*	46,23±2,12*	60,23±1,33*	69,68±1,48**	59,77±1,24**	68,43±1,54**
Area, mkm^2	89,73±2,44**	99,29±4,14**	95,11±3,12**	98,93±4,07**	174,82±6,77*	199,31±4,65**	169,72±4,83**	193,23±4,61**
Length, mkm	16,32±0,41**	19,43±0,55*	17,22±0,44*	19,32±0,57*	18,62±0,73	18,93±1,33*	17,72±1,06	18,87±1,23*
Width, mkm	8,61±0,51*	9,02±0,71*	8,91±0,54	8,95±0,72	17,78±0,43*	18,61±0,22**	16,77±1,36*	18,43±0,42**

* - validity p>0,05; ** - validity p>0,001.

CONCLUSIONS

Preliminary conclusion can be made that zeolite nanoparticles from Vangin, Kulikov and Lyutog deposits which are situated in the Far East of Russia reduce the consequences of burn injury in the skin. According to our data zeolite from Kulikov deposit has the least positive effect. This can be probably explained by the "needled" structure of the crystalline lattice. In general, the state of the burnt wound after zeolite applications showed the greater maturity of granulation tissue and greater functional viability of the fibroblasts than the patients' burnt tissue treated with "Levomekol" ointment applications. According to the literature (8, 9), the mechanism of zeolites action is to adsorb wound toxins, to add microelements in the wound, to normalize homeostasis in the injured area due to the catalytic properties of the crystal lattice. The problem of using zeolites in burning surgery requires further study. This work was supported by the Foundation for Assistance to Small Innovative Enterprises in science and technology (program U.M.N.I.K.) and RFBR grant 09-04-90781-mob_st.

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