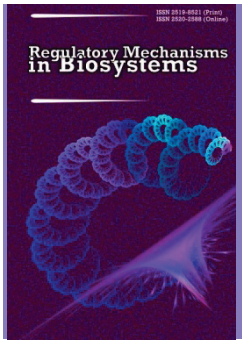

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Efficacy of extracts from cryopreserved placenta on third-degree burns in rats

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Human placenta extracts have anti-inflammatory, antioxidant and wound-healing properties, so they are promising drugs for the treatment of wounds of various origins, including burns. Cryopreservation methods are widely used to preserve the biological activity of placental drugs for a long time. The aim of this work was to study the effect of low-temperature storage of the placenta on the regenerative properties of its extracts. Fragments of freshly obtained placentas were cooled by immersion in liquid nitrogen and stored at –196 °C for 6 months. The placenta was warmed in a water bath at 37 °C. The effect of low-temperature preservation of the placenta on the ability of its extracts to positively affect the wound healing process was studied in a model of thermal burn of III B degree in rats. The effectiveness of wound treatment with extracts from cryopreserved placenta was evaluated by planimetric and histological methods at 3, 7, 14, 21 and 28 days after the burn. The activity of antioxidant enzymes in the serum of animals was also determined. superoxide dismutase activity was assessed by inhibition of adrenaline autooxidation in carbonate buffer, catalase activity was assessed by the degree of inhibition of ammonium peroxide formation. It has been shown that the treatment of burns with extracts from cryopreserved placenta helped to accelerate the regeneration processes and the rate of wound healing. The formation of granulation tissue was detected on the 7th day of treatment with extracts, and on the 14th day in the control. The area of burn wounds during treatment with extracts probably differed from the control starting from 14 days after application of the burn. It was found that the dynamics of recovery of catalase activity after burns is probably higher on the 7th day of treatment with extracts. The obtained data testify to the high efficiency of application of placenta stored at low-temperature for the purpose of obtaining extracts from it with preservation of regenerative properties.

Keywords: low temperature storage; human placenta extracts; burns; dynamics of wound healing; catalase; superoxide dismutase.

Introduction

The use of low temperatures, on the one hand, promotes more efficient release of biologically active substances into the extraction solution, and on the other it ensures the preservation of high viability of various cryopreservation objects, including cells and tissues of fetoplacental origin (Ananian et al., 2019; Nikulina et al., 2019; Prokopyuk et al., 2020). Central to such objects is the placenta itself, because as a non-classical but highly active endocrine gland, it purposefully influences the development and formation of all human organs and systems before birth. The placenta is a rich source of biologically active substances used in the treatment of a wide range of diseases (Adani et al., 2019; Nagae et al., 2020; Ghoneum & El-Gerbed, 2021). In particular, placental drugs are effective in regenerative medicine (Phonchai et al., 2020; Nejad et al., 2021; Sheehy et al., 2021).

Modern methods of placenta cryopreservation provide not only time for quality and thorough examination of biological material, but also the possibility of using natural components of placental tissue with preserved morpho-functional characteristics after long-term period of storage (Likhitskyi & Goltsev, 2019; Svyridyuk et al., 2020). Significant clinical efficacy of cryopreserved human placenta extract in the treatment of various pathological conditions has been shown (Goltsev & Yurchenko, 2013; Pelypenko & Shepitko, 2021). There is a large number of examples of successful use of cryopreserved placental extracts in gynecology, namely in the correction of urogenital disorders in women, in the immunization of pregnant women, in the treatment of diabetes during pregnancy, as well as in the treatment of chronic fetal hypoxia (Grischenko & Yurchenko, 2011).

Burns are a very common skin injury. Depending on the depth of thermal damage, burns are divided into three main categories: superficial burns, which affect only the epidermis, partial-thickness burns, in which both the epidermis and dermis are affected, and burns of full-thickness skin (Guo et al., 2020). Deep thermal burns are characterized by the development of general and local disorders that lead to a complex of interconnected processes in all body systems (Vaghardoost et al., 2019; Sterling & Lombardi, 2021). Burn wound healing is a complex process consisting of several continuous stages (inflammation, proliferation, tissue remodeling) (Guo et al., 2020). Excessive activation of lipid peroxidation processes, increased activity of free radical processes, which leads to the formation of oxidative stress in the burn wound and nearby areas of skin (Bhatia et al., 2016; Babu & Babu, 2018) is essential. The development of oxidative stress is accompanied by a decrease in the body's natural antioxidant defenses. The process of lipid peroxidation in the body is counteracted by the antioxidant system, the most important enzyme components of which are superoxide dismutase (SOD; EC 1.15.1.1) and catalase (CAT; EC 1.11.1.6), the combined action of which provides detoxification of potentially harmful oxygen reactive substances, i.e. anion-radical superoxide and peroxide. However, not only the imbalance of antioxidants and prooxidants, but also the violation of the ratio of enzymes to antioxidants can lead to increased oxidative stress due to the additional ge-

neration of reactive oxygen species. Their excess promotes the development of necrosis in the burn area and increases the level of lipid peroxidation (Sumsuzzman et al., 2020). Therefore, special attention should be paid to treatments aimed at balancing redox homeostasis and antioxidant status in skin burn injuries. Promising in this regard is the use of preparations of human placenta, in particular its extracts (HPE) (Pogozhykh et al., 2018). The aim of this work was to study the regenerative properties of extracts from cryopreserved placenta in the treatment of full-thickness burns in rats.

Materials and methods

The studies were performed on Wistar white male rats weighing 150–170 g in the vivarium of the Institute for Problems of Cryobiology and Cryomedicine of the National Academy of Sciences of Ukraine. All the experimental interventions and slaughter of the animals were performed in accordance with the requirements of the Bioethics Committee of the Institute for Problems of Cryobiology and Cryomedicine of the NAS of Ukraine, agreed with the Directive of the European Parliament and the Council of the European Union from 22.09.2010.

HPE obtained from placentas weighing 400–600 g (40 weeks gestational age, n = 6) were examined. Fragments of freshly obtained placentas were frozen in plastic bags to –196 °C by immersion in liquid nitrogen and stored for 6 months. Rewarming was performed in a water bath at 37 °C. Fragments of cryopreserved placenta were thoroughly washed from mucus and blood with isotonic NaCl solution, which was changed several times. The amniotic and chorionic membranes were separated from the washed placenta with scissors, following which the placenta was ground into 3 x 2 cm fragments, which were immersed in a vessel with saline in 1:5 ratio of placenta and solution (v/v) and stirred for 2–3 minutes. The supernatant was drained and a fresh portion of saline was added, and the procedure was repeated 3–4 times. Saline was added to the washed pieces of placenta in a volume ratio of 1 : 1 and homogenized with a high-speed homogenizer RT-1 U4.2 (Odessa Experimental Factory of Laboratory Medical Equipment, Ukraine) for 3 minutes. The homogenates were kept for 12 hours at +4 ºC, centrifuged at 3000 g for 15 minutes. The supernatant was collected and passed through a 0.45 μm membrane filter (Millipore Corp. Carrigtwohill, Ireland). The filtrate is an aqueous-salt extract of the placenta (Rozanova et al., 2011).

The effect of placental cryopreservation on the regenerative properties of its extracts was studied by their effect on the wound healing process in the model of thermal burn of III B degree in rats (Kornienko et al., 2019). To inflict burns on the outer surface of the thigh, the skin was trimmed and epilated in advance. After anesthesia, thermal contact (local) burns were performed with a device that contained a thermostat and a metal plate with an area of 2.5 x 2.5 cm, which was heated to 200 °C. The time of skin contact with the device was 10 seconds.

Treatment of burns was carried out by applying HPE (0.5 mL) to the wound two times a day, the wound surface was not isolated. The effectiveness of HPE was monitored by planimetric and histological methods on days 1, 3, 7, 14, 21 and 28 after the burn, the process of wound healing was assessed photographically (Chang et al., 2011). A ruler with a minimum scale of 1 mm was placed on the burn site and photographed with a Nicon D 5100 digital camera, the expansion of the obtained images was 1920 x 1080. Total wound area was calculated using a computer software ImageJ 1.52a. Body surface area in rats was calculated according to the Meeh formula in the modification of Gilpin (1996), the percentage of damaged area was determined by the formula described by (Hosseinimehr et al., 2010).

To assess the morphological changes of epidermal and dermal tissues, its reparative ability after thermal burns on days 3, 7, 14, 21 and 28 of the experiment after anesthesia, the animals were decapitated. Biopsy (1.5 x 0.5 cm) containing the central part of the burn wound and its edge with areas of intact skin adjacent to the defect area was taken from the wounds of the animals. The obtained skin fragments, no larger than 1.5 cm^3 in size, were fixed in a 10% neutral formalin solution for 2–10 days, after which they were washed daily from the fixative in water. Next, the selected pathological material was dehydrated in 70%, 80%, 90%, 96% and absolute ethyl alcohol for 20–24 hours each. Material was compacted with xylene (up to 1 hour), then xylene-paraffin (up to 40 minutes) at a temperature of 37 °C in a thermostat and paraffin at a temperature of 56 °C (up to 3 hours). Next, the pathological material was filled with paraffin, paraffin blocks were prepared. After solidification in cold water with ice, they were cut with a rotary microtome. The obtained sections were straightened in distilled water (+40 °C) and pasted on the prepared slide, after drying and dewaxing stained with hematoxylin-eosin (Ehrlich's hematoxylin) according to the general rules in pathohistological practice (Vaghardoost et al., 2019). Microscopy was performed on a microscope Achioscop-40 (Carl Zeiss) with a photographic nozzle.

For biochemical analysis the serum of intact animals (n = 8), was used, which determined the activity of SOD by inhibiting the autooxidation of adrenaline in carbonate buffer, pH = 10.53 (Misra & Fridovich, 1972) and the activity of CAT by the degree of inhibition of the formation of ammonium peroxides (Korolyuk et al., 1988). Spectrophotometric studies were performed with spectrophotometer Pye Unicam SP 8000 (Pye Unicam Ltd, UK). In the group of rats that were not subjected to thermal burns (intact group), no significant changes in the activity of SOD and CAT of blood were registered during 2 weeks of the experiment. The relative content of SOD and CAT as a percentage of the respective control indicators was also measured.

Results were statistically processed using the software package Statgraphics Plus version 2.1 for the OS Windows (Manugistic, Rockville, MD, USA). Independent studies were performed three times, after which the arithmetic mean value was calculated. The results are presented as the arithmetic mean ± standard deviation. Differences between the values in the control and the experimental groups were determined using ANOVA, where the differences were considered reliable at $P < 0.05$ (taking into account Bonferroni correction).

Results

Severe burn injury caused necrosis of all layers of the injured skin area with the attachment of adjacent tissues, which led to the formation of burns of III B degree (deep dermal burn, long-term healing by type of scarring, Fig. 1). The burns were uneven and spotty. In all rats, the injured skin was dry with a thin whitish-yellow scab, the epidermis did not exfoliate, underwent coagulation necrosis, and the animals were lethargic. At the end of the first day, the animals had a thickening and compaction of the scab.

On day 3, loose inhomogeneous surfaces with convex and drooping areas of yellowish-brown colour, cracks with serous exudate and blood impurities were observed in the group of control animals. Some wounds had areas with a pronounced serous-purulent exudate. In animals after treatment with HPE there were relatively clear contours of the wound edges. The surface of the damaged area was dry (excluding two animals, which had wet white areas of the wound), partially covered with a bumpy dark brown crust, there was the formation of a scab of dark purple colour.

In the burn area (Fig. 1) namely, the wound, in the upper part of the wound is a mass of necrosis with coagulation of all layers of the epidermis, papillary layer of the dermis, in the deep layers of the dermis a typical inflammation develops with a pronounced vascular reaction and leukocyte infiltration, in addition the zone of secondary lysis of fabric comes to light. On the periphery of the wound there are folded epithelial layer, leukocyte infiltration, vascular reactions (Fig. 2). There are also areas with purulent inflammation complicated by the microflora.

When calculating the surface area of the wounds, it was found that three days after the thermal burn in the control group and the group of animals treated with HPE, there were some increases in the area of the burn wound (Table 1). This is due to the fact that areas of the skin surface, which on the first day after the burn were bluish, then became a zone of necrosis.

The results of visual studies showed that on the 7th day after the burn the edge of the wounds in experimental animals were not in contact. In all rats, areas with microscopically defined necrosis turned into a necrotic scab of brownish-brown colour, firmly fixed with adjacent tissues, but there were differences in the appearance of the wound and the condition of the scab in groups. In 70% of animals in the control group, the wound surface was covered with a thick scab with protruding hair shafts, and in

rats with a thin scab on the wounds areas of purulent colour were observed with bloody exudate on the edge of the wound. When using HPE for the treatment of animals, the wound surface was mainly covered with a well-organized dense scab, fairly separated from the surrounding tissues, which protruded above the surface of the intact tissue. In 20% of animals, the onset of scab rejection was observed.

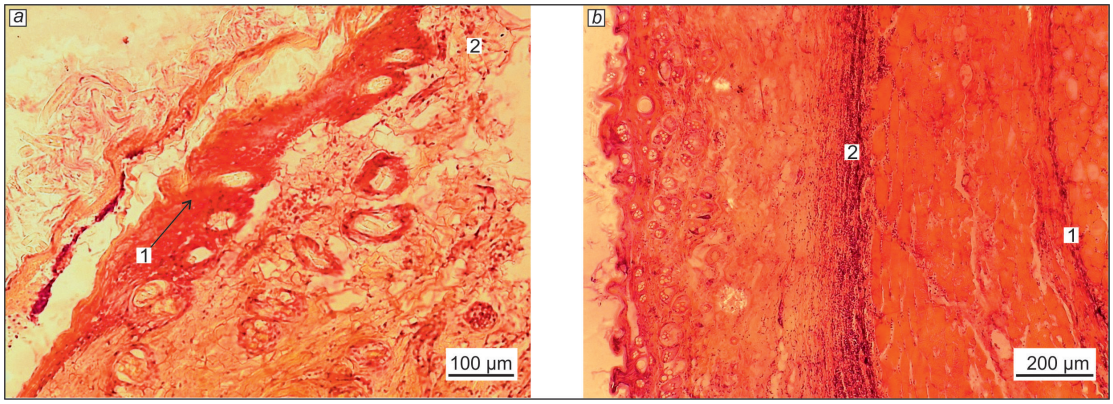

Fig. 1. Histological changes in skin of rats for first day after burn of III B degree:
1 – deep necrosis, *2* – acute inflammation, leukocyte infiltration; hematoxylin and eosin stain

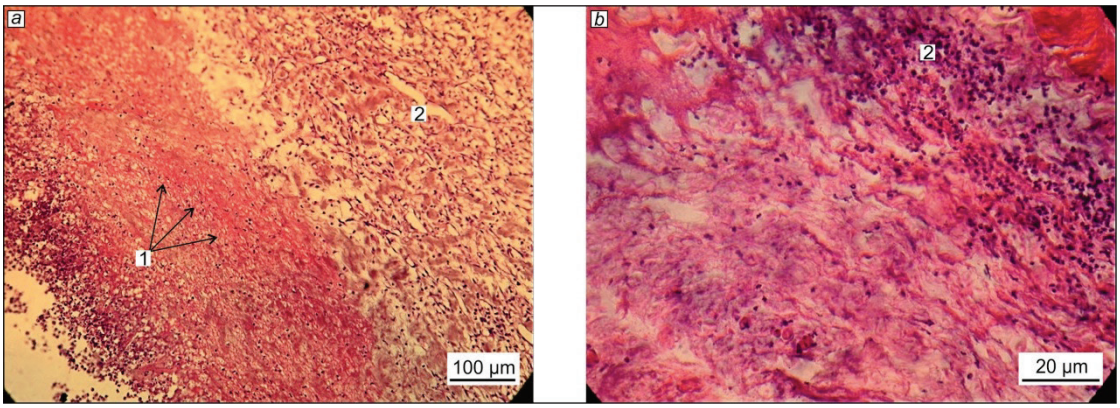

Fig. 2. Histological changes in skin of rats on the third day after burn of III B degree:
1 – formation of fibrinous tissue scab, *2* – leukocyte infiltration; hematoxylin and eosin stain

Table 1
Surface area of burn wound (mm^2) in skin of rats after burn of III B degree in control (without treatment) and after treatment with extracts from cryopreserved human placenta (HPE, x ± SD, n = 8)

Groups of animals	Number of days after burns					
	1	3	7	14	21	28
Control	319 ± 23	338 ± 26	316 ± 25	269 ± 14	75.7 ± 3.4	10.8 ± 1.5
HPE	312 ± 22	326 ± 27	300 ± 24	219 ± 13*	28.0 ± 3.1**	0.9 ± 0.3**

Note: significant differences compared to the control: * – P < 0.05, ** – P < 0.01.

On the 7th day of the experiment, there was an acute inflammation with fibrinous exudation that permeates all layers of the dermis (Fig. 3). At the border of the wound, we found hyperplasia of the marginal epithelium and its derivatives. There was healing by secondary tension. When HPE treated there was revealed marginal epithelialization of the wound, the proliferation of epidermal combs in the thickness of the dermis (Fig. 4). There were foci of proliferation of granulation tissue, pronounced vascularization (small capillaries were found) in the dermis. Visual studies of thermal burns of rats treated with HPE on day 11 corresponded to the second phase of the wound healing process. It is during this period that the wound is filled with newly formed granulation tissue, which is an important element in the process of reparative regeneration of the burn wound.

On the 14th day after the burn, the difference in the healing of burn wounds between the control and the experimental group of rats was more pronounced. The size of the wound surface generally decreased (Table 1), but a thick scab partially remained. Analysis of macrophotographic images showed that 60% of control rats had a thick scab with protruding hair shafts. In two rats, the scab was higher located over the level of intact tissue, the edges of which were uneven and wet with the smell of manure. Wounds with a partially absent scab and wet or dry granulation tissue of bright red colour were also observed. Histological research revealled places of formation of a scar with focal hyalinization (Fig. 5). After treatment with HPE in two rats on the 14th day, on the periphery of the burn wound granulation processes developed with a partially rejected scab (without clear boundaries); two wounds were covered with moist granulation tissue with minor remnants of scab; 50% of rats had a wet pink or bright red surface after scab rejection. The absence of a solid thick scab facilitated the migration of cells on the surface of the wounds. Scar, mature granulation tissue formed and the phenomenon of active epithelialization with foci of fibroblast proliferation developed after HPE treatment (Fig. 6) in contrast to the control group.

The antioxidant system of rats was assessed up to 14 days after the burn. Modeling of grade III B thermal burns led to a decrease in the activity of antioxidant enzymes in the blood of animals. A day after the thermal burn the activity of SOD, the main enzyme of specific antioxidant protection of the body, decreased relative to baseline by 1.3 times. The activity of CAT being an enzyme containing heme and leads to the decomposition of hydrogen peroxide into oxygen and water, on the first day after the burn decreased relative to baseline by 2.9 times, which reflects the development of oxidative stress (Fig. 7). On the 3rd, 7th, and 14th days of the study, CAT activity increased, but did not reach the level of baseline values. On the 7th day after the burn, the growth of CAT activity was more intense in the group of animals treated with HPE (77.4% vs. 63.3%).

The nature of changes in these indices was reflected in their balance. As can be seen from the results of the assessment of the balance of antioxidant enzymes, the CAT/SOD ratio on the first and third days decreased significantly due to a decrease in CAT activity (Fig. 7), ie there was an imbalance in the antioxidant system. Considering the role of CAT in oxygenation processes, it can be argued that animal tissues are in the stage of hypoxia, which is characteristic of the 1st phase of the wound healing process.

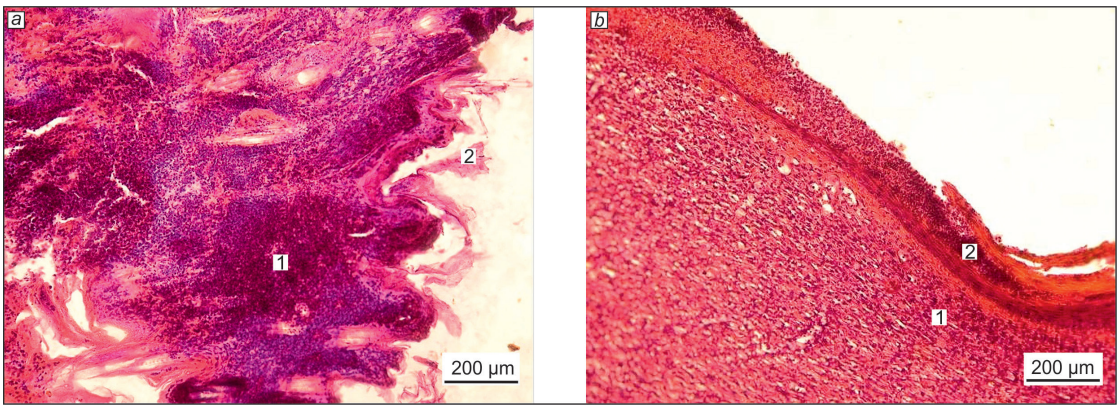

Fig. 3. Histological changes in skin of rats (control, without treatment) for 7 days after burn of III B degree:
1 – severe leukocyte infiltration, *2* – impregnation of exudate masses of necrosis, formation of fibrinous tissue scab; hematoxylin and eosin stain

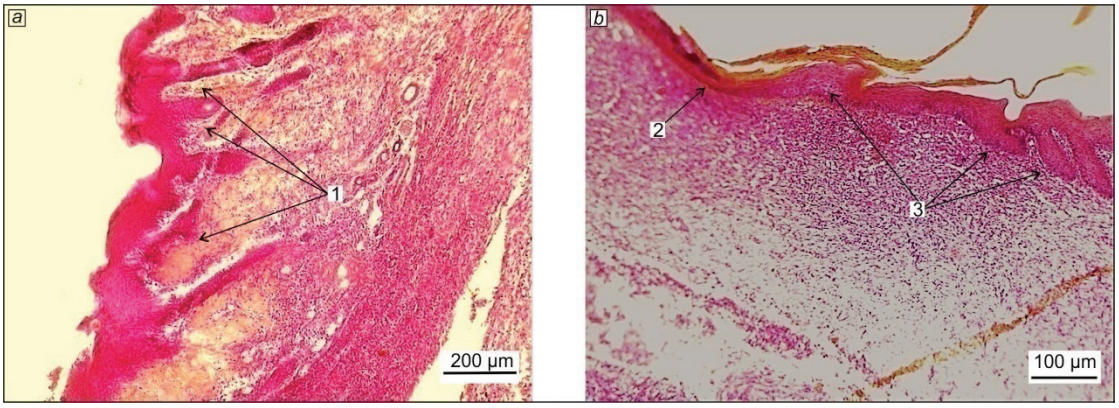

Fig. 4. Histological changes in skin of rats (after treatment with extracts from cryopreserved human placenta) on the 7th day after burn of III B degree:
1 – proliferation of granulation tissue, epidermal combs, *2* – boundaries of fibrinous tissue scab,
3 – hyperplasia of epidermis on wound edge, wound epithelization; hematoxylin and eosin stain

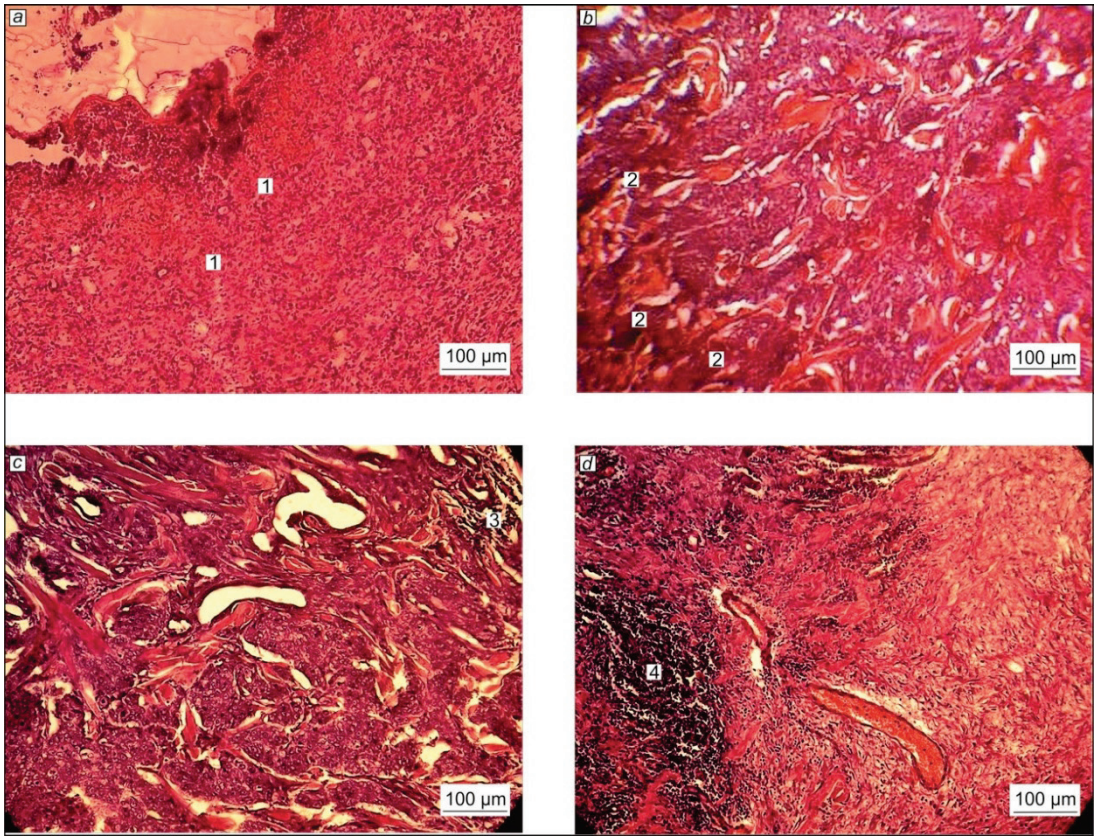

Fig. 5. Histological changes in skin of rats (control, without treatment) for 14 days after burn of III B degree:
1 – proliferation of granulation tissue, *2* – organization of necrosis masses with scar formation, *3* – scar with hyalinosis development,
4 – organization of necrosis masses, proliferation of macrophages, fibroblasts, hyperemia and paravasal edema; hematoxylin and eosin stain

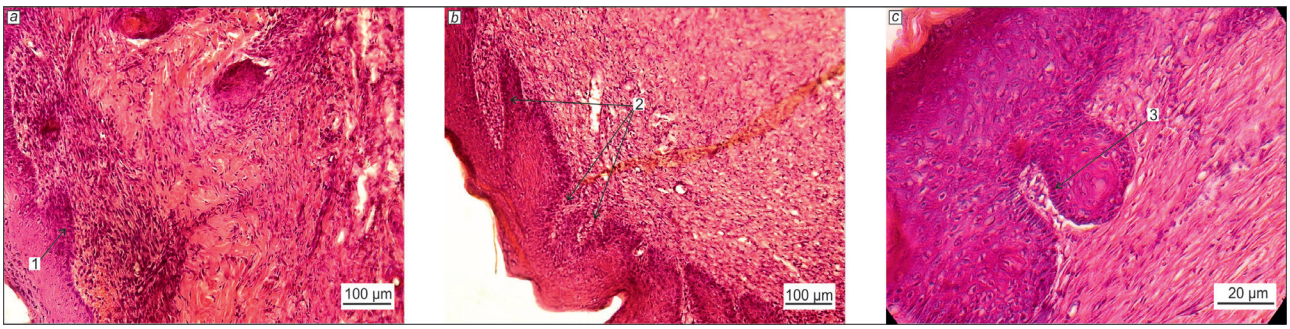

Fig. 6. Histological changes in skin of rats (after treatment with extracts from cryopreserved human placenta) on the 7th day after burn of III B degree: *1* – thickening of epithelium, formed granulation tissue, foci of proliferation of fibroblasts and histiocytes, *2* – processes of active epithelialization due to marginal hyperplasia epidermis, with formation of epidermal combs, *3* – hyperplasia of epithelium of the basal layer of epidermis, epidermal combs; hematoxylin and eosin stain

On the 7th day after thermal burns, the CAT/SOD ratio increases, but this occurs against the background of decreased SOD activity (44.9%) and increased CAT activity in the control group and the group of animals treated with HPE, relative to previous indicators (63.3% and 77.4%, respectively). This imbalance between the components of enzymatic antioxidant protection indicates the development of oxidative stress mainly due to the depletion of the ability of SOD to neutralize superoxide radicals. On the 14th day after the burn (the first and second phases of the wound healing process), there was a continued increase in the activity of antioxidant enzymes of both groups of animals and a tendency to restore the balance of the studied enzymes (Fig. 7).

Visual observation showed that on the 16th day after the burn, only in the group of rats treated with HPE, was complete clearance of the wound from necrotic scab observed. On the surface of the wound after the rejection of the scab a thin, shiny, pink granulation surface appeared.

On the 21st day, there was an independent cleansing of the burn wound from necrotic tissues in the control group, revealed hyperplasia of the epidermis and the phenomenon of epithelialization of the edge of the scar tissue (Fig. 8). In the presence of mature granulation tissue, foci of hair follicle proliferation were not detected. In the group treated with HPE, there was an active regeneration of all skin derivatives, manifested in dry, smooth granulation tissue, signs of epithelialization and the formation of different sizes and colours of scars. We can say that the healing of a burn wound on the 21st day after the burn occurs in the third phase of the wound process, which is reorganization of the scar and epithelialization of the wound. The reduction in the size of the defects is due to marginal epithelialization or wound contraction. The analysis of the area of the wound related to the total surface area of the animals revealed probable differences between the experimental and control groups of animals starting from the 14th day of the experiment (Fig. 9).

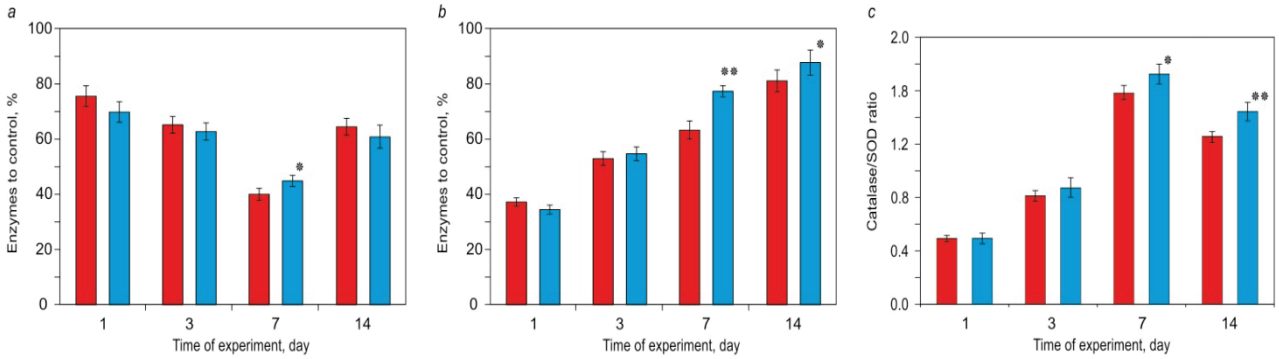

Fig. 7. Enzymatic activity in serum of rats after burns of III B degree in control (without treatment, red) and after treatment with extracts from cryopreserved human placenta (blue): the activity of SOD (*a*) and catalase (*b*) is related to the activity of the corresponding enzymes in healthy animals, the ratio of catalase / SOD (*c*); * – $P < 0.05$, ** – $P < 0.01$ compared with the control (in the corresponding day) using ANOVA taking into account Bonferroni correction ($x \pm SD$, $n = 8$)

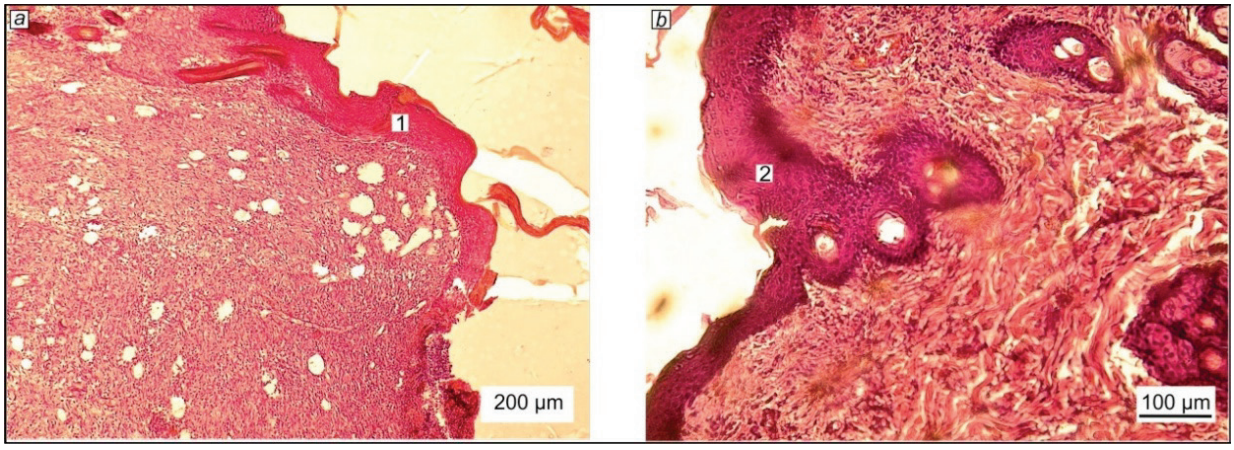

Fig. 8. Histological changes in skin of rats at 21 days after burns of III B degree in control (*a*) and after treatment with extracts from cryopreserved human placenta (*b*): *1* – hyperplasia of epidermis, epithelialization at scar tissue edge, *2* – active regeneration of all skin derivatives; hematoxylin and eosin stain

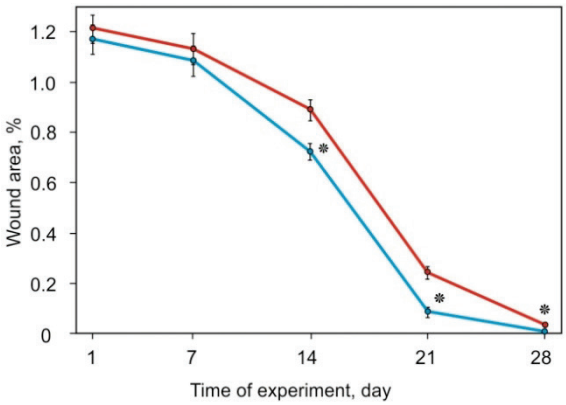

Fig. 9. Rate of wound healing in dynamics relative to entire body surface of rats after burns of III B degree in control (red line) and after treatment with extracts from cryopreserved human placenta (blue line): * – significant difference compared to control ($P < 0.05$, $n = 8$)

In the control group of rats on day 28 after the burn, the wound surface was covered with granulation tissue, scarring was observed. In the experimental group (HPE), the wound was completely healed and hair was restored. Epithelial hyperplasia was revealed (Fig. 10a) along the edge of dense scar tissue, along with proliferation of granulation tissue, in some – epithelialization with the formation of epidermal combs. For the group treated with HPE (Fig. 10b) there was a restoration of the skin, the formation of a group of follicles and sebaceous glands.

Discussion

In the area of damage from deep thermal burns on the first day, the wound healing process is in the first phase (Stone et al., 2018), which is accompanied by blood stasis in the capillaries and ischemia, resulting in weakened tissue respiration in cells of this area with subsequent death of epithelial cells of demarcation zone (skin necrosis) (Bhatia et al., 2016).

Anaerobic glycolysis leads to acidosis, which promotes the appearance of inflammatory mediators and rapid inactivation of antioxidant enzymes (Korać & Buzadžić, 2003). The primary source of cytokines and anti-inflammatory mediators in burns is the area of damage with increased migration of neutrophils (Baskaran & Yarmuch, 2000), which clean the damaged skin from the products of thermal decomposition of biomolecules with oxygen reactive substances. In the second phase (proliferation phase) of the wound process, when cleaning the wound from purulent-necrotic secretion and the formation of granulation tissue in it, extracellular and cellular proteolytic enzymes are of great importance. Therefore considering the cell elements involved into proliferation phase, it is advisable to accelerate angiogenesis, activate the synthetic function of fibroblasts to form granulation tissue, activate epithelialization of intact skin derivatives (Okur et al., 2020). To do this, it is advisable to use biologically active substances of natural origin (Vaghardoost et al., 2018; Thanapaul et al., 2020). Effective tissue stimulants are HPE. *In vitro* experiments in cell models have shown that HPE exhibits antioxidant and anti-inflammatory properties (Pogozhykh et al., 2008; Bobrova et al., 2018), and therefore can stimulate neoangiogenesis. The low-temperature effect does not have a negative effect on the biologically active components of the placenta and extracts from it have antioxidant, anti-radical and anti-inflammatory effects (Bobrova et al., 2018; Narozhnyi et al., 2018). The anti-inflammatory effect of HPE is accompanied by a decrease in platelet aggregation (Govorova et al., 2017) and therefore an increase in capillary permeability, which allows serum enriched with proteins to penetrate into the interstitial space. At the last stage of the wound healing process (third phase), when the epithelization of the wound and the reorganization of the scar begins, the drugs with a pronounced effect of stimulating cell growth are used. That is why HPE is most effective in the second and third phases of wound healing and eliminates skin defects. The wound-healing effect of HPE is due to numerous regulatory components and the fact that the placenta contains a number of growth factors (Sharma & Bhattacharyya, 2015; Pogozhykh et al., 2018). They cause immunomodulatory, anti-inflammatory, wound healing effect, promote cell proliferation and regeneration.

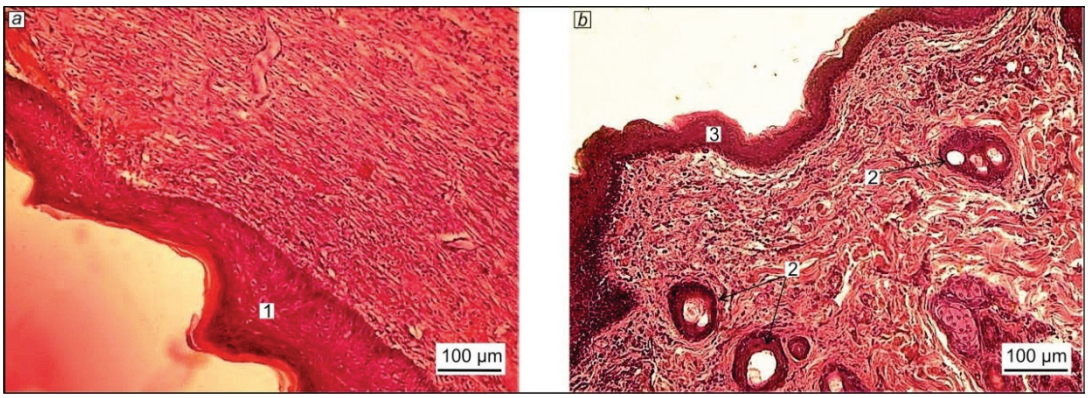

Fig. 10. Histological changes in skin of rats on the 28th day after burns of III B degree in the control (*a*) and after treatment with extracts from cryopreserved human placenta (*b*): *1* – skin epithelization, *2* – sebaceous glands, *3* – epidermis restoration; hematoxylin and eosin stain

In HPE, vasculoendothelial growth factor is manifested as a potential mitogen for vascular epithelial cells and has a significant effect on their permeability. It is known that epidermal growth factor accelerates the growth and regeneration of keratinocytes. Wound healing can also be the result of the activity of a peptide enzyme similar to fibronectin of the third type. This type of peptide has also been found in HPE (Nath & Bhattacharyya et al., 2007). Fibronectin forms a bridgehead in the wound, to which fibroblasts then migrate. Successful wound healing requires the presence of cytokines, the restoration of deficiency of which contributes to the effective treatment of chronic wounds. Cytokine TGF (transforming growth factor) is an immunoregulatory cytokine that has a great influence on intercellular connections and is actively involved in the regulation of cell growth and differentiation. Thus, at the stage of granulation there is an increase in the activity of cellular metabolism and the growth of fibroblasts, which begin to synthesize and produce a significant amount of extracellular collagen. The collagen content in the wound is regulated by many factors. Control of these processes provides a therapeutic opportunity to intervene in the process of wound healing and prevent pathological scar formation. Expanded necrosis can be reduced by general or local action on the wound, both HPE and other drugs from placenta.

Conclusions

Treatment of burn wounds with extracts from cryopreserved placenta helped to obtain a higher rate of healing, which was manifested in the acceleration of changes in the phases of the regeneration process: shortened period of cell infiltration and accelerated rate of granulation formation. Macroscopic differences in the acceleration of the wound healing process were observed by the 7th day after the burn in the form of rejection of the necrotic scab and the formation of granulation tissue. Thermal damage to the skin of rats was accompanied by a decrease in the activity of SOD and CAT in the blood, which indicates a decrease in antioxidant

protection of the body. The positive effect of HPE in the treatment of burns was manifested in a more intense increase in the activity of CAT by the 7th day of treatment.

The study was carried out within the framework of the state theme "Influence of cryopreservation of placenta and its water-salt extracts on antioxidant and anti-inflammatory action of extracts" (state registration No. 0116U003491).

The authors have no conflicts of interest to declare. All co-authors have seen and agree with the contents of the manuscript and there is no financial interest to report.

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