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Research Article

DETERMINATION OF THE MECHANISM OF HEMOSTATIC ACTION OF GEPROCELL IN AN EXPERIMENTAL MODEL OF **HEAT INJURY**

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ABSTRACT

In case of long-term non-healing wounds the skin defect persists for a long time, significant cicatricial and dystrophic changes occur in the surrounding tissues, there is no tendency for stable independent epithelialization. Therefore, surgical treatment of long-lasting wounds that do not heal has taken the leading place in pathogenetic therapy, and it is primarily autodermoplasty. Any wound with a diameter of more than 5 cm needs to be covered artificially with a flap or skin slices to achieve a lasting therapeutic effect and a good functional result. A wound can be closed with local tissue grafting: excision of the edges and the bottom of the wound, mobilisation of the edges and a dummy suture. In some cases the skin can be mobilised by means of a loosening incision. The wound can also be closed by applying secondary sutures with excision of the edges and the wound bed (late secondary suture).

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KEYWORDS

Autodermoplasty, regeneration, combustiology, Geprocell.

Introduction

In combustiology there are several scientific studies on the development and application of implants with a blood stopping effect, following the reactive regeneration processes epitheliocytes in the injured liver after autodermoplasty on a global scale. To this end, there is an urgent need to develop and apply in clinical practice biosynthetic and synthetic film coatings that carry hemostasis, reduce bleeding during necrectomy and autodermoplasty, and carry the epithelialisation process in the injured area. Among them, a carboxymethyl cellulosebased bioparam with a high styptic effect is considered, and there is the possibility of using it as a topical medication. In this regard, research studies are being conducted to evaluate the efficacy of necrectomy and autodermoplasty in patients with severe thermal insufficiency, to create a hemostabilizing coating with high therapeutic efficacy. In deep burns, development of specially designed biodegradable

hemostabilizing coatings in autodermoplasty is an urgent task [1,10].

PURPOSE OF THE STUDY

Determination of the mechanism of hemostatic action of geprocell in an experimental model of heat injury

MATERIALS AND METHODS

The topical styptic is a polymeric implant in powder and film form with bioactive styptic properties. It is a homogeneous film of transparent to crisp colour, its solubility in water at 20 °C is 10 mg/l, it is well soluble in most organic solutions and its melting point is 220 °C. It decomposes rapidly in a stable alkaline environment at 5-7 RN and is much more stable in an acidic environment [7,9].

A model of 18 male white rats weighing 180-210 g (Rattus norvtgicus f.domesticus) was carried out in the experimental laboratory of the

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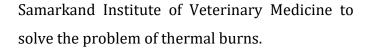








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As requested by the Declaration of Helsinki, we approached animals in connection with humanity and shaped the burn with light efir drugs. We developed a methodology to simulate deep thermal burns using a hot fluid of 90-93.80°C in rats. General anaesthesia is administered using Halothane steam, and the rat is fixed abdominally on the operating table under sterile conditions. The fur under the hind part has been mechanically cleaned. The skin is cleaned again from a surface 0.5 cm wider than the surface where the burn is caused. The area of the burn is approximately straightened and the back part should be about 20%. The burn area of an animal weighing 180-210 g was 10-12 cm2 or about 2.5-3x4 cm [5, 7, 8].

The study was conducted on the surface after necrectomy during autodermoplasty in experimental rats. Eighteen animals took part for the experiment. The study was conducted by dividing the experimental calamuschlars into 2 groups.

- Animals of the control group 1 underwent postnecrectomy and skin transplantation without hemostatic heprocellin preparation.
- In animals of study group 2, post-necrectomy surgery and skin autodermoplasty with the hemostatic preparation Heprocelantine were performed.

In the morning, the experimental animal was taken to the operating theatre. With Halothane steam, the area formed by the burn injury was prepared for incision using special modes of the ro-6 machine under inhalation anesthesia (first under bell, later under masked anesthesia). From the spruce-covered wound, the edges were cleared of wool to a distance of 0.5 cm. Then, taking into account the size of the cut area, the area of the right thigh was scraped and cleaned. Necrectomy of healthy tissue was performed concomitantly with capillary haemorrhage. Hemostasis in the control group (n=9) was performed by gauze transfusion and coagulation in the active areas. Hemostasis in the main group (n=9) was carried out by coagulation of MERGEL vessels in the areas of active bleeding and hemostasis was performed using a hepatocellular implant. In the experimental group of animals, we performed hemostasis through a hemostatic

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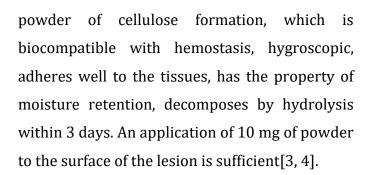








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During this time, after necrectomy, the skin was cut through the entire skin layer along an acute pathway from the prepared area to obtain skin of an appropriate size for the resulting wound in advance. The skin was placed on a special surface and sutured with clasps. Subcutaneous fat was then removed using a tear lens and the skin muscles were incised to the level of the hair follicles. A necrectomy was performed and a skin incision was made on the surface of the incision. The edges of the incision were sutured to the edges of the skin wound using a separate 4/0 profile suture. Due to the viscosity of the bloodstopping powder, the donor graft strengthened and did not require special fasteners.

After dermoplasty, the animals were left in their cages on their normal diet. For the first 2 days after surgery, 0.5 ml of the anaesthetic drug ipobrufen was added to drinking water. By various methods, thermal burns in laboratory animals are known to be simulated by a process using special devices. But a series of popular models associated with the production of the device has not been produced.

We have developed a methodology to simulate deep thermal burns using hot liquids in rats. General anaesthesia is carried out with Halothane steam and the rat is fixed abdominally on the operating table under sterile conditions. The fur under the hind part is mechanically cleaned off. A further 0.5 cm wide surface is cleaned from the surface, which is called the skin burn. The area of the burn shall correspond to 20% of the back, which is roughly the shape of the dogleg. The burn area of an animal weighing 180-210 g was 10-12 cm2 or about 2.5-3x4 cm.

The device has a hemispherical shape with a receptacle and has 3 openings. No. 1 opening is intended to make contact with the animal's body, the size and configuration of which is called the burn zone and is suitable for the back of the animal. This opening is hermetically sealed with a thin rubber plug. Hole no. 2 is a 1000s and is intended for the injection of liquid into the warmth reservoir, no. 1 is 50-100 ml, depending on the size of the opening. Hole no. 3 is hermetically connected to a 0.8 cm support tube,

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the end of which is connected to a vessel for draining water. The unit is used in the same way:

Hole no. 1 is applied to the back of a sleeping animal in a state of anaesthesia. At the agreed time, boiling water is applied to the tank with the tank through opening #2. No. 3 ensures a constant water temperature of 90-93.80 °C at the point of contact of the animal's body. By controlling the temperature of the water at the point of contact with the skin, it is possible to accurately record exposure times, create burn wounds similar in area and degree of storage of the sealant. A grade 3 burn was achieved with a contact temperature of 9±1 sec over time.

Results: During the experiment, we did not need to create a protective covering against contact with the surface of the jarochete, so that the burn injury in the animals could end up in vivo. The cage in which the animals are kept is treated with pre-disinfection solutions. For the first 3 hours after the formation of the burn, the animals did not feel any effect due to the continuing evasion effect of halothane. On the following days, we added ipobrufen to the animal's drinking water for 3 days.

For a short time after the formation of the burns. the animals were in a state of postnarcotic intoxication. Loss of orientation, walking time was shaky and they could not stand well on their feet. Within 2 hours of the operation, the animals regained consciousness. They protected the areas where the burn injury had occurred and retreated to a corner. It is noted that rapid breathing and pulse rate are increasing. The jungle is not smooth. It should be noted that in the postoperative period it was observed that animals given anaesthetics combined with saline and physiological solution in the abdominal cavity to correct water-electrolyte metabolism lost consciousness very slowly, came out of anaesthesia strongly and had difficulties in recovering their breath. In 6 rats administered saline (up to 10 ml) and anesthetics: 50% analgin 0.3 or 0.1% - 0.1 dimedrol were observed after 3 hours in 3 units, 3 animals were given oxygen which was withheld for 2 hours due to severity of their condition, 2 of them died the next day after the experiment. 1 piece of squid was healthy. The animals in the following experiment were not injected with fluid into the abdominal cavity and no anaesthetic drugs were given as injections.

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The animals were given ipobrufen from 0.5 to 200 ml of water for 2 days as an analgesic. A state of death was not observed after the formation of burns in the animals of this group.

Observation of the condition of the animals revealed the following (Table 1). Twenty hours after third-degree burns on 6% of the body surface, the animals were drinking water, averaging their movements. They were often kept in a corner of the cage and in a relaxed posture. Contamination of the burn surface was not allowed. The colour of the wound in the area of the burn was smooth and elastic, and on examination it was observed to be fluid and retained. No blistering or outflow of fluid was observed.

Within 3 days, the animals became much more fawning, the burn area began to be covered in black spots, and black spots appeared in the intermediate areas. The burn area intensified and a demarcation line in the form of hyperemia began to appear on the feel of the arang. And in one part of the animals, the contamination manifested itself as the surface of the wound was moist and thickened its fur over the burn area. The animals became much more mobile, wanting water and food. The weight loss was up to 20%, depending on the initial condition.

The animals remained quite active for 7 days and began to be well on water and food. The burn area is covered with a dark brown scab, not hard, not elastic. The burn area was significantly reduced by 80% compared to the original appearance. Almost all animals showed scab infestation in the form of a mucous cut, which was slightly separated from the granulation wound, covered by a cortical plague. No scab detachment was observed with acute bleeding from the wound. Weight loss is up to 15%, depending on the initial condition.

Within 14 days after rutting, the animals are quite mobile, drinking water and moving much more actively. A weight gain of 5-10% was noted compared to the 3rd day after the burn. The resulting burn area was closed by a rigid scab with signs of contamination. The area is reduced by up to 30%. In most animals, the wound scab is taken without haemorrhage, under which detachable tissue with slight detachment is found, covering the surface with a film.

On the 21st day after rutting, the animals are sufficiently mobile. There was no increase in

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weight compared to day 14. The area of the scorch decreased to 40% and became oblong, starshaped. In most animals, the fluff that had detached from the wound was replaced by new

fluff, more tightly adhering to the wound, difficult to remove, with signs of petechial haemorrhage.

1 table Dynamics of rat body weight (g) M±m in the main and control groups at different times of the experiment

	Test time	Animal weight dynamics (g).		Р
		basic	control	
1.	As long as it does not burn out	198,44±1,72 (π=35)	196,42±1,86 (π=18)	>0,05
2.	Day 1	195,29±1,63 (π=35)	193,15±1,79 (π=27)	>0,05
3.	Day 3	191,20±1,60 (π=35)	189,42±1,79 (π=27)	>0,5
4.	5th day	189,68±1,48 (π=35)	187,19±1,70 (π=27)	>0,05
5.	7th day	189,11±1,49 (π=35)	186,53±1,69 (π=27)	>0,05
6.	Day 12	190,02±1,57 (π=35)	187,03±1,75 (π=27)	>0,05
7.	day 20	195,11±1,80 (π=10)	179,66±2,45 (π=10)	<0,005

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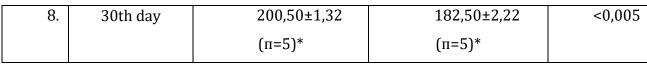








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* - value for start-up time

On the 30th day after the burn, the animals become active. The tendency for contraction on the burn surface continued and began to take on an irregularly elongated shape. On the surface of the wound - a rumen is visible on which the bark is moving and forming. The weight of the animals was stable. At day 40 all animals were healthy. Most animals developed an irregularly shaped scar, which was 20% of the original burn injury. In 30% of the rats, the bark of the Jarohat halicum was retained in some areas of the wound, tightly, well attached to the wound. The weight of the animals returned to its original state. On day 60 of the experiment, the in all animals was completely finished with the formation of a long white scar, no hair emergence was observed.

In laboratory rats, natural adhesion processes of deep skin burns with the addition of microbial flora were observed in all cases from a practical point of view. Histological studies as well as observation of animals after formation of an experimental skin burn showed the onset of infection from day 3 after wound surgery. On day experimental animals showed signs of

contamination in the wound of 50%. Limited burns (up to 20% of the body surface) and clinical signs of normal animals were very rarely seen. The animals remained active, moving freely around the cage, eating and drinking water. The moment of inspection was not observed in the majority of animals where the scab was covered with Pugal, heavy, detached from the wound. In 10% of cases there was a rash on the surface of the scab - a rash and bruise, a slight separation from the wound. There were histological signs of infiltration with neutrophilic leukocytes. Day 3, an important sign of burn injury is the appearance of a demarcation line, which appears as a light reddish-coloured frame around the perimeter of the burn. At a much earlier time, such a boundary between the burn area and healthy tissue was not macroscopically formed.

For 4 days or more after the occurrence of a deep skin burn, almost all experimental animals showed signs of contamination of the wound under the scab, and necrectomy against a background of infection prevented the graft from undergoing dermoplasty due to the risk of

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migration. The animals in the experiment continued the process of reducing the existing skin defect under the scab. A gradual reduction in the volume of the scab led to a narrowing of the area by pulling the scab slowly out of the marginal areas. Depending on the size of the injured area and the degree of contamination of the wound, the healing process lasted up to 60 days. With the completion of the inflammatory and reparative processes, the final stage ended with the formation of an elastic, almost imperceptible stretch scar, often in the form of a broken line or zigzag. The scars in our studies were not observed with the hypertrophic or kelloid transformation inherent in laboratory animals of this type.

Thus, to accelerate the healing of burn injuries in laboratory rats, it is optimal to perform autodermoplasty with neurectomy within 3 days after the burn, when contamination in the wound is minimal. Due to the obvious development of the process, demarcation some difficulty determining the boundary of the skin incision was eliminated. The depth of the cut was determined according to the degree of tissue bleeding and reached the muscle. Late dermoplastic process was observed with contamination and migration of the skin graft.

In our experimental studies, a non-cractomy day 3 procedure was performed: the extent of deep skin burns was up to 20% of the body surface, and no parenteral fluids had to be sent as the animals naturally replenished the fluids lost on their way out In our experiments, no death was observed from a 20% burn on the posterior surface. The fact that the animals were in vivo after the burn formed and the ligament was not closed on the 4th resulted in an almost 100% contamination of the wound in 24 hours, in which case it was not possible to perform primary plasty after necrectomy. The day after surgery, the animals became a regular asset, trying to retaliate against the surgical intervention areas. We added ipobrufen to the water in order to anaesthetise the wounds after the operation for 2 days. The protective ties were not put on.

Day 9 after autodermoplasty. Viable area of grafted skin can be seen along the right contour of the wound. The left quadrant is still covered with spruce, with part of the plantain partially necrotic. The 2 animals with partial skin necrosis also had a dense scab covering approximately 50% of the cuticle surface. Other signs of contamination are evident. The animals were active in the area of the lesion without restriction.

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2. there is complete necrosis and contamination on the skin of the animals. The jar is completely covered with uneven, rough spruce.

Infiltration, reddening of the jarohat chetes is observed. Injuries are free of streaks. The animals are very active.

9 animals in the control group had complete completion of skin autotransplantation by partial reduction (up to 30%) in the defect area with complete repair of the defect in 2 units (30%). The skin sutures were removed on day 8. An increase in wool was observed at the autograft site. A thin elastic scar with no signs of hypertrophy or inflammation remained within 7 days of the donor area.

Of the 9 animals in the post-necrectomy and autodermoplasty group. complete disappearance of the defect was observed on day 7 after the dermoplasty, and a partial necrosis was observed on day 14. No state of death was observed.

In the animals of the main group, the same method of operation was performed on day 3 after the formation of deep skin necrosis in the dorsal and jagged areas, using a new device and boiling water. A difference in the operative

intervention in rats of this group was the use of the haemostatic preparation Heprocel to stop the blood flow after incision of the skin and subcutaneous structures with necrosis of the fascia and muscles. After application of a bloodstopping film up to 50 µm thick from the cellulose formation, bleeding completely stopped and the surface of the jar began to take on a glossy appearance due to the film's adhesion to the surface. Skin retrieval and treatment on the skin was the same as in the control group. It was noted that the adhesion of the skin tissue to the wound underneath was much better than in the control group when the voruet defect was closed. No additional fasteners or restraints were applied during the postoperative period. The skin was sutured with a donor area similar to that obtained in the control group animals.

Day 3 after surgery. In the main series of operated animals, positive dynamics of the cuticle bite were observed. 5 The condition of the rats is good and they are active. In constant movement in the cage. Youngs are soft, shiny. Skin tone is normal in colour, soft elastic. The condition of the skin sutures is good. There are small patches of spruce on the suture line, up to 1 cm wide. No separation from the wound. The donor site is also clean, the

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sutures are in good condition and show no signs of contamination. Against a background of good general condition, 1 rat in the same series developed a scab, occupying less than 2% of the grafted skin, 4 to 50 mm wide on the plantar surface of the skin along the suture line. The skin is soft, supple, and of normal colour, where it is not covered with scab. There are no signs of contamination. There were signs contamination in the donor skin area in all control rats.

Conclusion

The technology of performing recently developed autodermoplasty with the hemostatic agent Hepocel has significantly improved the results of surgical treatment of deep burns. Moment autodermoplasty after necrectomy allows the wound to close quickly by rubbing the hemostatic agent Hepocel into the local marotaba, resulting in rapid and complete (100%) skin healing. After an initial tangential necrectomy, the application of Hepocel at the time of autodermoplasty ensures complete hemostasis and leads to better revascularisation during free skin squatting. At early necrectomy and application of Heprosel early animal activity and weight gain is

manifested by a rapid attenuation of the clinical signs of burn intoxication.

Application of Heprocell makes it possible to carry out autodermoplasty simultaneously or delayed with early necrectomy, it is convenient to change direction of burn disease and to stop its course, time of restoration of skin integrity, duration of hospital treatment, number of infectious complications and reduction of the percentage of burn disease are reduced.

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