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# Application of platelet autocryolsate in the treatment of mine-blast wounds – case report

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### Abstract

Mine blast injuries are characterized by multiple blood vessel damage, as well as damage to nerve fibres, muscles and skin. Infection is an additional aggravating factor. One treatment method is cell therapy. A promising direction of this technique is platelet autocryolysate (PAC), an abiotechnological product based on the oligopeptides and growth factors obtained from the patient's blood platelets. A patient was treated who had suffered a long-term non-healing mine blast wound of the soft tissues of the left leg. Two secondary surgical debridements were performed, followed by vacuum therapy. The inflammatory process intensified and was complicated by phlebothrombosis of the deep veins of the leg. After two applications of PAC, the wound size decreased, the discharge disappeared, and the wound surface was covered with a dry, loose crust, under which active epithelialization occurred. Using PAC has a positive therapeutic effect in treating long-term, non-healing blast wounds of the tissues.

# Key words

infection, growth factors, blast wounds, platelet autocryolysate

# INTRODUCTION

During military operations on the territory of Ukraine, the number of wounded is rapidly increasing which requires the development and implementation of new, effective methods of treatment and rehabilitation. In modern military operations, the most significant part of combat wounds are mine-explosive wounds – 70%–75% of the total [1]. The consequences of these injuries most often (60–65%) are injuries to the extremities, with the lower extremities affected three times more often.

Blast trauma is a multifactorial traumatic injury resulting from the detonation of explosives. The trauma develops due to the shock wave, wounding projectiles, and exposure to gases, flames, and toxic substances. The combination of mechanical, thermal, and chemical damage, differences in the composition of explosives, distance, and external conditions, determine the significant polymorphism of blast trauma. Blast trauma is often complicated by shocktraumatic, burn, or haemorrhage.

Mine blast injuries are characterized by multiple damage to blood vessels and nerve fibres, and significant damage to muscles and skin [2]. In most cases, infection is an additional aggravating factor [3]. These factors can influence the woundhealing process and delay it for an extended period [4]. Platelet-rich plasma (PRP) is a treatment method that can significantly improve regenerative processes [5–7]. A promising direction of this technique is the use of platelet autocryolysate (PAC), a biotechnological product based on the oligopeptides, and growth factors obtained from the patient's blood platelets [8,9]. The basis of PAC is obtaining cryolysate of high-quality human platelets from a minimum volume of the patient's blood, which is guaranteed to have a sufficient clinical effect.

The presented clinical case demonstrates the effectiveness of PAC in treating a long-term non-healing battle wound. The case is based on medical documentation provided by one of the authors of the report (A. Barkovsky), the surgeon who treated the patient.

# **CASE REPORT**

**Patient B.** On 18 November 2022, a 44-year-old male, received in combat a mine blast wound which caused predominant damage to the soft tissues of the left shin. Primary surgical treatment (removal of foreign bodies from the wound, excision of dead tissue, haemostasis) was provided directly in the medical unit. Two days later, the patient was taken to the hospital for inpatient treatment. On 2 December, secondary surgical treatment was performed to remove an accumulation of pus. Subsequently, the wound was treated daily with chlorhexidine solution, and amoxil was used for antibacterial therapy at 1,000 mg twice a day. Although some

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positive dynamics were observed, the wound did not heal. After two weeks, the inflammatory process intensified with increased local oedema.

On 21 December 2022, the decision was made to perform another operation for secondary surgical treatment of the wound, followed by vacuum therapy. In subsequent treatment, an intravenous infusion of 100 mg pentoxifylline in 100 ml of 0.9% sodium chloride solution was used daily to improve peripheral circulation. Physiotherapeutic procedures included darsonval and laser therapy, once a day, alternately.

Despite the treatment, complete wound healing by secondary intention did not occur. The inflammatory process intensified again, cyanosis and an increased pattern of subcutaneous veins appeared in the wound area, oedema increased, and the patient complained of bursting pain in the wound area.

On 28 December 2022, due to complicated phlebothrombosis of deep veins of the leg, secondary sutures diverged. The decision was made to continue conservative wound management until it healed. Drug therapy was enhanced by the addition of dioflan to improve capillary blood flow, one tablet in the morning and evening, and cardiomagnyl to prevent secondary thrombus formation – 150 mg daily. Physiotherapy was represented by magnetic therapy and pressotherapy procedures once a day. As a result of the ineffectiveness of this treatment, a long-term non-healing trophic skin ulcer was formed.

Eight months later, on 2 September 2023, the wound was 4x2 cm in size. The edges were undermined and moderately oedematous. There were compactions with pronounced cicatricial changes. The bottom of the wound was covered with pale pink granulations and fibrin films. There was also a moderate serous-purulent discharge on the dressings, and signs of peripheral thrombophlebitis (Fig. 1). The patient had an unstable dermatogenic flexion contracture in the knee joint: extension up to 140 degrees, accompanied by pain in the wound area characterized by 5/10 on the VAS.

On 2 September 2023, due to the ineffectiveness of therapy, the patient was offered treatment with PAC. Following the 1975 Declaration of Helsinki, written informed consent was obtained from the patient.



Figure 1. Condition of the wound before using PAC

Treatment. The production of platelet autocryolysate for treatment was carried out using the following method [10]. The patient had blood drawn with an anticoagulant (sodium citrate) in a ratio of 9:1, followed by double centrifugation. After the first centrifugation at 2,500 rpm for 10 minutes, the plasma was separated from the red and white blood cells, and the precipitated fibrin removed. After a second centrifugation at 3,200 rpm for 3 minutes, the platelet pool sediment is removed from the platelet-depleted plasma. When the amount reached about 1x109 platelets/ml, the resulting precipitate was filtered and collected in a cryovial frozen in liquid nitrogen to obtain a lysate product containing growth factors. Before use, the resulting lysate product is thawed to 37 °C. After preliminary sanitation of the bottom of the wound and surrounding skin with a chlorhexidine solution, the patient was given a one-time injection of 4 ml of platelet autocryolysate. A total of 3 procedures were performed with an interval of seven days The wound was dressed with an aseptic gauze bandage every other day, treated with chlorhexidine, and pantestin ointment applied.

The wound was visually assessed a week after the first procedure (Fig. 2).



Figure 2. Condition of the wound after first PAC injection

The wound decreased in size to 1.7x0.5 cm, and the perifocal oedema and exudation from the bottom of the wound decreased. The edges of the wound became softer, and the discharge was replaced by serous in small quantities.

A week after the third procedure, the size of the wound decreased to 0.3x0.5 cm, the discharge disappeared, and the wound surface was covered with a dry, loose crust under which active epithelization was taking place. The surrounding skin was without inflammation or swelling, cellulite and thrombophlebitis were absent, and the severity of cicatricial changes in the skin edges minimal (Fig. 3). Movement in the knee joint resumed in full and without pain.

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Figure 3. Condition of the wound after third PAC injection

# DISCUSION

In recent years, interest in the use of cell technologies in orthopaedics and traumatology has been growing constantly worldwide. Research conducted has revealed the prospect for using regenerative medicine, in particular cell therapy, to treat a wide range of diseases of different origins. As already mentioned, in most cases platelet-rich plasma (PRP) is used to attract SCs into the regenerative process [11, 12].

Platelet-rich plasma is understood as the patient's autologous plasma, in which the concentration of platelets is increased several times, compared to the initial values of peripheral blood by stepwise centrifugation [13]. The primary active agent of PRP is the platelet – nuclear-free blood cells – which, in addition to participating in haemostasis, play an important role in regeneration processes, releasing so-called growth factors (GFs) – complex peptide compounds contained in alpha granules – into damaged tissues, and can stimulate cell growth and division [14, 15].

It has been proven that platelets, in addition to growth factors, contain hundreds of other bioactive proteins in their structure that play a crucial role in homeostasis and induce the involvement of leukocytes in the regeneration process. Upon contact with the extracellular matrix at the site of injury, platelets secrete a wide range of chemokines that attract leukocytes, including CXCL4 and CXCL7, the most abundant platelet chemokines. In addition, platelets promote the migration and fixation of leukocytes to the site of inflammation. However, due to mechanical and chemical influences, a significant part of the cells is destroyed during platelet-rich plasma preparations, thus the final product contains fragments of destroyed cell membranes, metabolic products, intracellular biologically-active substances. Using platelet autocryalizate, which ensures the preservation of a high concentration of platelet growth factors, can avoid the influence of these undesirable factors [16].

Wounds with considerable necrotic damage to skin and muscle structures can result not only from explosions, but also from exposure to high temperatures. Firefighters record such injuries when extinguishing forest fires [17, 18]; this applies especially to wounds caused by the so-called 'fire whirlwind', which is typical for forest fires [19]. In this regard, the proposed method of treating such wounds becomes even more relevant.

## CONCLUSIONS

Using PAC has a positive therapeutic effect in the treatment of long-term, non-healing blast wounds of the skin. The proposed technique guarantees the production of platelet growth factors from a given number of cells; after a single blood draw, the required amount of individualized cryolysate preparations can be obtained for further use.

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