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A case of autologous thrombofibrin clot use in a patient with post-burn persistent corneal erosion

E.V. Fedoseeva*[1], E.V. Chentsova[1], N.V. Borovkova[2], I.N. Ponomarev[2], A.O. Petrova[1], Yu.A. Pavlenko[1]

[1]*Moscow Helmholtz's Research Institute of Eye Diseases, 14/19 Sadovaya-Chernogryazskaya St., Moscow 105062 Russia;*

[2] *N.V. Sklifosovsky Research Institute for Emergency Medicine, 3 Bolshaya Sukharevskaya Sq., Moscow 129090 Russia*

*Correspondence to: Elena V. Fedoseeva, Postgraduate of the Department of Trauma and Reconstructive Surgery, Moscow Helmholtz's Research Institute of Eye Diseases, e-mail: dr.elfed23@gmail.com

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The purpose. *Using clinical observation, to study the feasibility of applying a coagulant and to assess its efficacy in the treatment of patients with post-burn erosion of the cornea.*

Material and methods. *The treatment was carried out in the Department of Trauma and Reconstructive Surgery, the Helmholtz Moscow Research Institute of Eye Diseases, Russia. Based on clinical test results, the patient, was diagnosed with post-burn persistent erosion of the cornea with sub-total cornea conjunctivization and: limbal stem cell deficiency (LSCD) in the right eye. The keratoprotective therapy conducted for several months turned ineffective. In order to activate the cornea regeneration process, the erosion site was covered with autologous thrombofibrin clot that had been obtained from the stabilized blood. Following a two-stage centrifugation of patient's stabilized blood, the plasma with the platelets were collected into a round-bottomed test-tube; and the chloride calcium activator was added.*

The clot was formed in the thermostat at a temperature of 37 C. The patient was given a local anesthesia, and the thrombofibrin clot was placed on the cornea surface and covered with *an amniotic membrane. The membrane was fixed with a circular stitch along the limbus of the cornea.*

Result. *The patient was followed-up and showed positive changes over time in the form of a diminished erosion area at day 5 and a complete corneal epithelialization at day 14.*

Summary. *Our methodaimed at enhancing the reparative and regenerative processes in the cornea, reducing swelling, accelerating epithelialization, expanding transparency, and improving the optic characteristics thanks to less intensive opacity formation showed a high efficacy.*

Keywords: cornea, burn, persistent erosion of the cornea, platelet count, clot, autologous plasma

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PRP, platelet-rich plasma

PCE, persistent corneal erosion

Introduction

The problem of blindness and poor vision is one of the most important socio-economic and ethical problems in the world. According to WHO

(2006), corneal blindness accounts for 4–5% among all causes of blindness in world population. According to the data from regions of the Russian Federation for 2010, the patients with corneal blindness accounted for 5.9% of all blind and visually impaired people in Russia. By its structure, the corneal blindness in the Russian Federation is associated with corneal ulcers in 9% of cases, with scars and corneal opacities in 21%. Among all traumatic eye injuries, the patients with eye burn disease account for 6%-38.4%. One of the most severe complications of burn disease is the persistent corneal erosion (PCE) leading to corneal ulceration and perforation.

The main principle of the PCE treatment is the enhancing of the reparative and regeneration processes in the cornea, as well as improving the epithelium adhesion to the underlying stroma. A conservative treatment includes topical and systemic drugs, such as moisturizing eye drops, hyperosmotic agents, matrix metalloproteinase-9 inhibitors, corticosteroids, doxycycline [1, 2]. Therapeutic and bandage contact lenses can also provide symptomatic relief and promote the corneal epithelium healing. A surgical treatment includes superficial keratectomy using a razor blade fragment or a diamond knife, phototherapeutic keratectomy [3–8], amniotic membrane transplantation [9], the use of cultured buccal epithelial cells [10, 11].

However, despite the existing approaches to the treatment, the problem of persistent erosion remains relevant, which leads to the search for the most effective method of treatment for post-burn cornea erosion. One of these methods implies the use of growth factors contained in platelet-rich plasma (PRP). It is known that the PRP contains over 1000×10^9 cells/L, i.e. 3-4 times more platelets than the whole blood. And human platelets, in turn, have a pronounced regenerative effect due to the alpha-granules being a

natural depot of growth factors and promoting the reconstruction of the damaged epithelium [12-14] due to enhancing the processes of proliferation, migration and differentiation of epithelial cells [15, 16]. Thus, the use of PRP in the treatment of corneal injuries is promising. In addition, the PRP has proven efficient in the treatment of ocular surface diseases, such as severe corneal xerosis, ocular manifestations of the graft versus host disease, persistent epithelial defects, neurotropic keratopathy [16-25]. J. Alio et al. reported that autologous PRP eye drops could be used to successfully treat corneal ulcers after laser-assisted in situ keratomileusis [26, 27]. Also, PRP has been used to treat patients with recurrent herpetic corneal erosion [28]. The experience of using an autologous thrombofibrin clot in patients with deep corneal ulcers and descemetocele to promote the scar tissue formation and to perform subsequent optical keratoplasty has been described [29].

However, this technique has been used in a small sample size of patients, and the standard treatment algorithm has not yet been developed.

The purpose of this study was to evaluate the feasibility of using a thrombofibrin clot and to assess its efficacy in treating patients with post-burn cornea erosions.

Material and methods

Patient V., born in 1975, had the following diagnosis on hospital admission "Post-burn persistent corneal erosion with subtotal conjunctivization, limbal stem cell deficiency *(LSCD)* of the right eye.” A standard examination included: visometry, tactile palpation for approximate intraocular pressure assessment, biomicroscopy with fluorescein-stain of the sample. The medical history: From the patient's words, the burn with boiling water occurred during everyday life activities. He was hospitalized to a local

hospital at the place of residence where he received a long-term conservative treatment. After 3 months from the injury the patient referred to Moscow Helmholtz's Research Institute of Eye Diseases with the complaints of the lack of pattern vision, foreign body sensation, mucous discharge from the eye.

On admission it was noted that Vis OD was pr. l. and certae OS was 1.0. At examination, the palpebral fissure was narrowed. The injection of conjunctival vessels and corneal conjunctivization with dilated, crimped vessels were noted. There was an erosion of 5.5x5.0 mm in diameter, with an uneven surface in the center of the cornea (Fig. 1, 1a). The anterior chamber was of medium depth, the pupil being in the center, the details were behind the flair. Deep structures were not visualized.

Fig. 1. Status on admission. Anterior segment biomicroscopy. Fluorescein staining illustrates the erosion

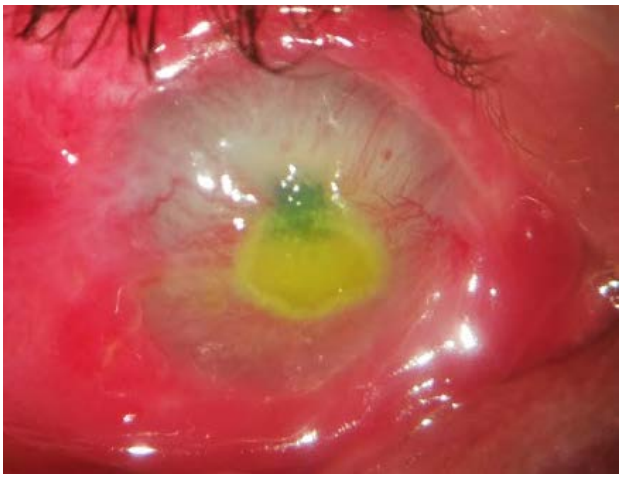

Fig. 1a. Status on admission (intraoperatively). Fluorescein-stained erosion of 5.5x5.0 mm in size

Considering that the patient had been given a long-term (3 months) reparative therapy with keratoprotectors that gave no definite effect, the decision was made to use a thrombofibrin clot. The informed consent to using an autologous thromfibrin clot was obtained. The patient also received anti-inflammatory, detoxifying, and reparative therapy.

On the eve of the surgery, the patient's blood in the amount of 20 ml was taken from the cubital vein and collected into tubes containing EDTA-K2 anticoagulant. Keeping the sterility, the blood was transported at a temperature of 22–24 C to the Sklifosovsky Research Institute for Emergency Medicine for preparing a personalized biograft. After a two-stage centrifugation in compliance with the aseptic and antiseptic rules, the plasma with the platelets nearest to the erythrocytes was selected, avoiding the lymphocyte layer. PRP in the amount of 3 ml with a platelet concentration of 1370×10^9/L was obtained; 1 ml of the obtained PRP was placed in a round bottom tube and 50 μl of 10% calcium chloride were added for the activation. After mixing, the tubes were incubated at 37° C in a

thermostat until the thrombofibrin gel was formed. As a result of constriction, the gel separated into a clot and a serum.

Results and Discussion

The resulting clot was placed on the surface of the cornea, and then covered with an amniotic membrane on top. The membrane was fixed with a circular suture to the episclera along the border of the limb; autologous serum was injected in the 4 quadrants along the limb (Fig. 2). After that, a temporary lateral blepharorrhaphy was performed.

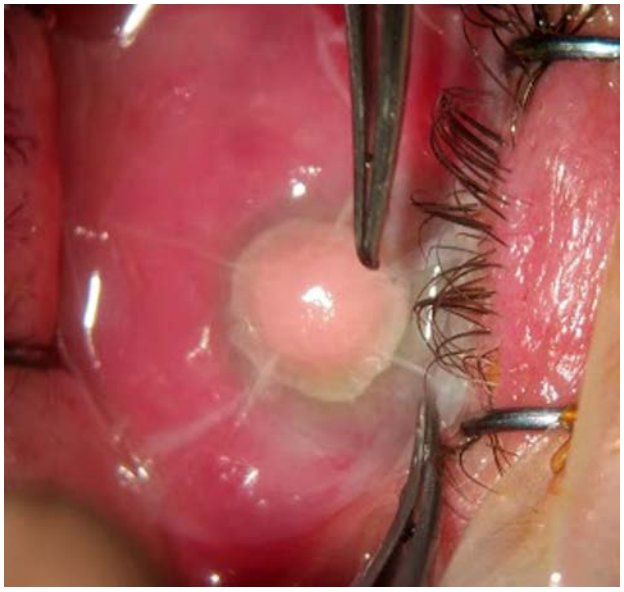

Fig. 2. The clot is placed on the cornea surface and covered with amniotic membrane

After 2 days, the clot flattened fitting the cornea under the amniotic membrane (Fig. 3).

Fig. 3. The clot is placed on the cornea surface and covered with an amniotic membrane. Status after 2 days post-surgery

On day 5, the sutures on eyelids were removed. The amniotic membrane and the thrombofibrin clot residues were removed to assess the epithelialization. At examination, an even fluorescein staining was seen. The erosion area reduced to 4.0x4.0 mm, acquiring more regular boundaries (Fig. 4).

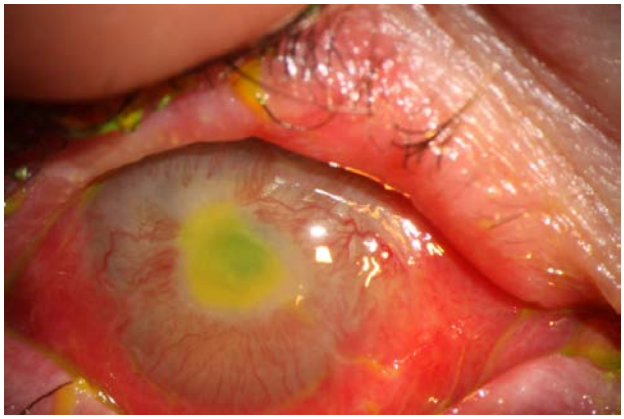

Fig. 4. The patient's eye on day 5 after surgery. The positive changes observed over time

Considering the positive dynamics, but an incomplete healing of the defect, the procedure was repeated. The patient was discharged home for an outpatient follow-up with the recommendations to visit the clinic after 2

weeks. At re-examination after removing the sutures from the eyelids and removing the amniotic membrane residues from the cornea, a decrease in the eyeball injection was observed; the cornea was completely epithelialized; there was no corneal defect at fluorescein staining. Vis OD at counting fingers near the face was eccentric (Fig. 5).

Fig. 5. The patient's eye after 2 weeks post-surgery. Staining with fluorescein demonstrates no corneal defect

Conclusion

The treatment method we used was aimed at enhancing the reparative and regenerative processes in the cornea, at reducing the inflammatory response and improving optical properties due to less intense opacification, slower conjunctivization, showed a high efficacy in a patient with persistent corneal erosion resulted from a burn disease.

Further studies of this method are necessary to develop algorithms for treating patients with corneal lesions of various origins.

References

1. Dursun D., Kim M.C., Solomon A., Pflugfelder S.C. Treatment of recalcitrant recurrent corneal erosions with inhibitors of matrix metalloproteinase-9, doxycycline and corticosteroids. *Am J Ophthalmol*. 2001;132(1):8-13. PMID:11438047

2. Hope-Ross M.W., Chell P.B., Kervick G.N., et al. Oral tetracycline in the treatment of recurrent corneal erosions. *Eye (Lond).* 1994;8(4):384-388. PMID:7821456 DOI:10.1038/eye.1994.91

3. Buxton J.N., Fox M.L. Superficial epithelial keratectomy in the treatment of epithelial basement membrane dystrophy: a preliminary report. *Arch Ophthalmol.* 1983;101(3):392-395. PMID:6600925

4. Soong H.K., Farjo Q., Meyer R.F., Sugar A. Diamond burr superficial keratectomy for recurrent corneal erosions. *Br J Ophthalmol*. 2002;86(3):296-298. PMID:11864887

5. McLean E.N., MacRae S.M., Rich L.F. Recurrent erosion: treatment by anterior stromal puncture. *Ophthalmology*. 1986;93:784-788. PMID:3737123

6. Kim S.Y., Ko B.Y. Evaluation of anterior stromal puncture using Nd:YAG laser for refractory recurrent corneal erosion. *J Korean Ophthalmol Soc*. 2015;56(3):331-8. DOI:10.3341/jkos.2015.56.3.331

7. Choi M., Jung J.W., Seo K.Y., et al. Comparison of Nd:YAG laser versus conservative management in the treatment of recurrent corneal erosion. *J Korean Ophthalmol Soc.* 2015;56(5):687-93. DOI:10.3341/jkos.2015.56.5.687

8. Forster W., Grewe S., Atzler U., et al. Phototherapeutic keratectomy in corneal diseases. *Refract Corneal Surg.* 1993;9(2 Suppl):85-90. PMID:8499387

9. Makarov P.V., Kugusheva A.E., Slepova O. S., et al. On persistent erosions of the corneal transplant. part 2. *Russian ophthalmological journal*. 2015;2:41-46. (In Russian).

10.Egorova N.S., Chentsova E.V., Borovkova N.V., et al. Experimental repair of deep corneal defects using a bio-construct comprising a collagen type I matrix loaded with buccal epithelial cells. *Transplantologiya. The Russian Journal of Transplantation*.2017;9(3):226-235. (In Russian). DOI:10.23873/2074-0506-2017-9-3-226-235

11. Egorova N.S., Chentsova E.V., Flora S.V., et al. Technologies of creation, evaluation of biocompatibility and safety of the collagen matrix in the bioengineering cell construction. *Russian Ophthalmological Journal*.2017;10(2):71-77.(In Russian).DOI:10.21516/2072-0076-2017-10-2-71-77

12. Nishida T., Nakamura M., Ofuji K., et al. Synergistic effects of substance P with insulin-like growth factor-1 on epithelial migration of the cornea. *J Cell Physiol.* 1996;169(1):159-166. PMID:8841432 DOI:10.1002/(SICI)1097-4652(199610)169:1<159::AID-JCP16>3.0.CO;2-8

13. Van Setten G.B., Viinikka L., Tervo T., et al. Epidermal growth factor is a constant component of normal human tear fluid. *Graefes Arch Clin Exp. Ophthalmol*. 1989;227(2):184-187. PMID:2785946

14. Gupta A., Monroy D., Ji Z., et al. Transforming growth factor beta-1 and beta-2 in human tear fluid. *Curr Eye Res*. 1996;15(6):605-14. PMID:8670763

15. Liu L., Hartwig D., Harloff S., et al. An optimised protocol for the production of autologous serum eyedrops.*Graefes Arch Clin Exp Ophthalmol*. 2005;243(7):706-714. PMID:15756576 DOI:10.1007/s00417-004-1106-5

16. Tsubota K., Goto E., Fujita H., et al. Treatment of dry eye by autologous serum application in Sjogren's syndrome. *Br J Ophthalmol*. 1999;83(4):390-5. PMID:10434857

17. Ogawa Y., Okamoto S., Mori T., et al. Autologous serum eye drops for the treatment of severe dry eye in patients with chronic graft-versus-host disease. *Bone Marrow Transplant*. 2003;31(7):579-83. PMID:12692625 DOI:10.1038/sj.bmt.1703862

18. Noble B.A., Loh R.S., MacLennan S., et al. Comparison of autologous serum eye drops with conventional therapy in a randomised controlled crossover trial for ocular surface disease. *Br J Ophthalmol*. 2004;88(5):647-52. PMID:15090417

19. Kojima T., Ishida R., Dogru M., et al. The effect of autologous serum eyedrops in the treatment of severe dry eye disease: a prospective randomized case-control study. *Am J Ophthalmol*. 2005;139(2):242-246. PMID:15733983 DOI:10.1016/j.ajo.2004.08.040

20. Tananuvat N., Daniell M., Sullivan L.J., et al. Controlled study of the use of autologous serum in dry eye patients. *Cornea*. 2001;20(8):802-806. PMID:11685055

21. Tsubota K., Goto E., Shimmura S., Shimazaki J. Treatment of persistent corneal epithelial defect by autologous serum application. *Ophthalmology*. 1999;106(10):1984-1989. PMID:10519596 DOI:10.1016/S0161-6420(99)90412-8

22. Poon A.C., Geerling G., Dart J.K., et al. Autologous serum eyedrops for dry eyes and epithelial defects: clinical and in vitro toxicity studies. *Br J Ophthalmol*. 2001;85(10):1188-1197. PMID:11567963

23. Young A.L., Cheng A.C., Ng H.K., et al. The use of autologous serum tears in persistent corneal epithelial defects. *Eye (Lond).* 2004;18(6):609-614. PMID:15184926 DOI:10.1038/sj.eye.6700721

24. Jeng B.H., Dupps W.J. Jr. Autologous serum 50% eyedrops in the treatment of persistent corneal epithelial defects. *Cornea.* 2009;28(10):1104-1108. PMID:19730088 DOI:10.1097/ICO.0b013e3181a2a7f6

25. Matsumoto Y., Dogru M., Goto E., et al. Autologous serum application in the treatment of neurotrophic keratopathy. *Ophthalmology*. 2004;111(6):1115-1120. PMID:15177961 DOI:10.1016/j.ophtha.2003.10.019

26. Alio J.L., Abad M., Artola A., et al. Use of autologous platelet-rich plasma in the treatment of dormant corneal ulcers. *Ophthalmology.* 2007;114(7):1286-1293.e1. PMID:17324465 DOI:10.1016/j.ophtha.2006.10.044

27. Alio J.L., Pastor S., Ruiz-Colecha J., et al. Treatment of ocular surface syndrome after LASIK with autologous platelet-rich plasma. *J Refract Surg*. 2007;23(6):617-9. PMID:17598582

28. Loshkareva AO, Maychuk D.Yu. Use of Platelet-rich Plasma in Patients with Chronic Corneal Erosions. *Modern Technologies in Ophthalmology*.2016;4:131-132.(In Russian).

29. Alio J.L., Arnalich-Montiel F., Rodriguez A.E. The role of "eye platelet rich plasma" (E-PRP) for wound healing in ophthalmology. *Curr Pharm Biotechnol.* 2012;13(7):1257-1265. PMID:21740369

Information about authors

Elena V. Fedoseeva, Ophthalmologist, Postgraduate of the Department of Trauma and Reconstructive Surgery, Moscow Helmholtz's Research Institute of Eye Diseases, ORCID: 0000-0002-8824-3562;

Ekaterina V. Chentsova, Prof., Dr. Med. Sci., Head of the Department of Trauma and Reconstructive Surgery, Moscow Helmholtz's Research Institute of Eye Diseases.

Natal'ya V. Borovkova, Dr. Med. Sci., Head of the Scientific Department of Biotechnologies and Transfusiology, N.V. Sklifosovsky Research Institute for Emergency Medicine, ORCID: 0000-0002-8897-7523;

Ivan N. Ponomarev, Cand. Med. Sci., Researcher of the Department of Biotechnologies and Transfusiology, N.V. Sklifosovsky Research Institute for Emergency Medicine, ORCID: 0000-0002-2523-6939;

Alena O. Petrova, Senior Researcher of the Department of Trauma and Reconstructive Surgery, Moscow Helmholtz's Research Institute of Eye Diseases, ORCID: 0000-0002-8888-6090;

Yuriy A. Pavlenko, Ophthalmologist, Postgraduate of the Ophthalmic Plastic Surgery and Ocular Prosthetics Department, Moscow Helmholtz's Research Institute of Eye Diseases.