CLINICAL AND EXPERIMENTAL
OPTOMETRY

RESEARCH

Autologous serum eye-drops and enhanced epithelial healing time after photorefractive keratectomy

Clin Exp Optom 2018; 101: 34–37 DOI:10.1111/cxo.12574

Hanife Tuba Akcam* MD
Metin Unlu[†] MD
Emine Esra Karaca[‡] MD
Hulya Yazici[§] MD
Bahri Aydin[¶] MD
Ahmet Murad Hondur[¶] MD

*Department of Ophthalmology, Cankiri State Hospital, Cankiri, Turkey
[†]Department of Ophthalmology, Erciyes University School of Medicine, Kayseri, Turkey
[‡]Department of Ophthalmology, Numune Training and Research Hospital, Ankara, Turkey
[§]Department of Ophthalmology, Sincan State Hospital, Ankara, Turkey
[¶]Department of Ophthalmology, Gazi University School of Medicine, Ankara, Turkey
E-mail: hanifetuba@hotmail.com

Submitted: 22 December 2016
Revised: 28 March 2017
Accepted for publication: 1 May 2017

Background: The aim of the study is to test whether use of autologous serum eye-drops can provide earlier epithelial healing following the application of photorefractive keratectomy.

Method: Sixty patients (60 eyes) underwent photorefractive keratectomy for myopia. Thirty eyes received autologous serum drops (Study group) while 30 eyes received conventional artificial tears (Control group) after photorefractive keratectomy. An 8 mm epithelial opening was prepared with the application of 18 per cent alcohol for 20 seconds. Photorefractive keratectomy was performed using ESIRIS excimer laser (SCHWIND, Kleinostheim, Germany) with an optic zone of 6.5 mm. Total duration of epithelial healing was monitored as the main outcome measure. The comparisons were done with chi-square test and independent samples t-test. Statistical significance was considered at $p < 0.05$.

Results: Preoperative myopic spherical refraction and ablation depths were similar in the study and control groups. The mean duration for epithelial healing was about one day shorter in the eyes receiving autologous serum compared to the eyes receiving conventional treatment (2.2 ± 0.25 days versus 3 ± 0 days, $p = 0.001$). All eyes achieved 6/7.5 or better uncorrected visual acuity in six months. In both groups, more than 90 per cent of eyes were within ± 0.50 D of emmetropia in 12 months. No significant difference was noted for the incidence of +1 haze.

Conclusion: Use of autologous serum drops reduces epithelial healing duration following surface ablation for two days.

Key words: autologous serum, cornea, photorefractive keratectomy, surface ablation

In the past 30 years, excimer laser therapy has become the major technique for the treatment of a wide spectrum of refractive errors.[1] The main procedures for excimer laser therapy are photorefractive keratectomy (PRK) which is also known as surface ablation, and laser-assisted *in situ* keratomileusis (LASIK). Both procedures create flaps before laser treatment and change the shape of the anterior central cornea by ablating (remove by vaporisation) a small amount of tissue from the corneal stroma. However, the flap in surface ablation is more superficial and epithelial whereas it is thicker and also includes some anterior stromal tissue in LASIK. Laser-assisted sub-epithelial keratectomy (or laser epithelial keratomileusis, LASEK) is a variant of PRK. While the corneal epithelium is removed and discarded with PRK, it is rolled to its original position at the end of surgery with LASEK.[2]

Among excimer laser techniques, LASIK remains as the main surgical technique due to fast postoperative recovery and patient comfort. However, LASIK carries risk for some unfavorable complications such as flap-related problems (for example, buttonholes, irregular flaps, free caps and flap dislocation) and risk of ectasia. As a result of those risks, the technique cannot be preferred by refractive surgeons for some classes of patients, particularly those who have thin corneas,[3] and who do contact sports.

Due to its suitability for those eyes in which LASIK surgery is risky and relative safety in terms of complications and technical simplicity compared to LASIK, surface ablation has remained as an alternative technique to LASIK. However, relatively slow visual recovery and early postoperative pain due to delayed epithelial healing remain the main disadvantages of surface ablation.[4]

The positive effect of autologous serum (AS) eye-drops on corneal wound healing, due to high concentrations of nutrients, growth factors and vitamins it harbors, has been demonstrated by many studies.[5–7] In a recent study, with the use of AS drops, we have achieved shortening of postoperative pain and epithelial healing after LASEK to three days, one day shorter than conventional duration.[8] In this study, we report the results of use of AS drops following PRK in an attempt to further shorten postoperative pain and accelerate visual recovery in surface ablation.

METHODS

Sixty patients (28 men and 32 women) with myopia were evaluated in the current study in the Gazi University Eye Clinic. Each patient received 20 per cent AS in artificial tears (Refresh Tears; Allergan, Irvine, California, USA) (Study group, n = 30) or conventional artificial tears (Refresh Tears; Allergan) (Control group, n = 30). The study was designed as double-blind style and the power of the test was 0.8 which

© 2017 Optometry Australia

means the model will reject a false null hypothesis with 80 per cent probability.

Using simple randomisation procedures, patients were randomly assigned to receive either AS or conventional artificial tear eye-drops. Written informed consent was obtained from all patients and the study was approved by the Gazi University local ethics committee. The tenets of the Declaration of Helsinki were followed throughout the study. Participants did not receive a stipend.

Inclusion criteria were 18+ years of age, stable refraction of at least one year, normal corneal topography, and a minimum central corneal thickness of 500 μm. Daily-wear soft contact lenses were removed at least one week prior to preoperative examination. Preoperative evaluation included medical history and complete ophthalmologic examination, including uncorrected visual acuity (UCVA), best spectacle-corrected visual acuity, manifest and cycloplegic refractions, anterior segment examination, applanation tonometry, ophthalmoscopy, corneal topography, pachymetry, and Schirmer testing. Patients with unstable refraction, dry eye, blepharitis, corneal disease, glaucoma, systemic diseases including infectious and collagen vascular diseases, diabetes, and topographical evidence of keratoconus, were excluded.

Whole blood (20 ml) was procured by venipuncture, centrifuged at 1,500 revolutions per minute (relative centrifugal force of 25.15 × *g*) for 10 minutes. AS drops were carefully prepared under a laminar airflow cabinet, and diluted with artificial tears (Refresh Tears; Allergan) to 20 per cent.[9] AS drops were prepared on the day of surgery and patients were asked to keep AS drops refrigerated at about 4°C.

Although all patients were treated bilaterally in the same surgical session, one eye of each was randomly selected for the study. Topical proparacaine 0.5 per cent was used to anaesthetise the eyes. A drape and lid speculum were inserted following the treatment of eyelids with 10 per cent povidone-iodine. In all eyes, an 8 mm alcohol well was placed centrally, and 18 per cent alcohol was applied for 20 seconds. After the epithelium was detached and discarded, a bare corneal surface with regular epithelial edge was achieved.

Laser ablation was performed using ESIRIS excimer laser (SCHWIND, Kleinostheim, Germany). Spherical ablations were performed according to manifest refraction without any reduction using the SCHWIND ORK-CAM aspheric profile. The optic zone diameter was 6.5 mm in all eyes with varying transition zones determined by SCHWIND ORK-CAM aspheric profile. Total ablation diameter did not exceed 8.0 mm in any of the eyes.

Once ablation was finished, the cornea was irrigated with chilled balanced salt solution, and a drop of moxifloxacin 0.5 per cent (Vigamox; Alcon Laboratories, Fort Worth, Texas, USA) was instilled. A cooled soft contact lens was placed over the cornea with sterile forceps. A drop of AS or artificial tears was instilled. The eyelid speculum and drape were removed.

The contact lenses were removed on epithelial closure. Patients were examined daily by the same observer early in the morning until epithelial closure. In eyes with incomplete but minor epithelial defects after contact lens removal on presumed epithelial closure, a new contact lens was placed and these eyes were examined in the late afternoon on Day 2. If the epithelium closure was observed in the afternoon of Day 2, total duration of epithelial healing was accepted as 2.5 days. All patients were examined after one, three, six and 12 months.

Postoperative medication until epithelial closure consisted of topical moxifloxacin 0.5 per cent (Vigamox; Alcon Laboratories) four times per day, and AS eye-drops (for Study group) or artificial tears (for Control group) (Refresh Tears; Allergan) every two hours. Topical moxifloxacin was discontinued, and AS eye-drops were changed to artificial tears after epithelial closure. Dexamethasone 0.1 per cent (Maxidex; Alcon Laboratories) was administered four times daily for one month followed by fluorometholone 0.1 per cent (Flarex; Alcon Laboratories) four times daily for another two to four weeks depending on refraction and haze level. All medications were discontinued after two months.

In daily visits, patients were asked to grade their level of ocular discomfort until epithelial closure as follows: Level 0, no pain; Level 1, mild pain; Level 2, moderate pain; Level 3, severe pain.

Postoperative haze was graded as follows: 0, completely clear cornea; +0.5, barely visible corneal opacity; +1, reticular subepithelial opacities not interfering with visibility of fine iris details; +2, punctate or coalesced subepithelial opacities with mild obscuration of iris details; +3, confluent subepithelial opacities with moderate obscuration of the iris and lens; and +4, dense opacities with complete opacification of the stroma.[10]

Statistical analysis was performed using SPSS 16.0 software (SPSS Inc., Chicago, Illinois, USA). Normality of all values was confirmed using Shapiro–Wilk test. The comparisons were done with the chi-square test for categorical variables, and independent samples t-test for continuous variables. Statistical significance was set at $p < 0.05$.

RESULTS

The mean age was 31.53 ± 10.18 years in the Study group versus 32.01 ± 7.56 years in the Control group ($p = 0.841$). The mean preoperative myopic spherical refraction was −3.42 ± 1.72 D (range: −1.25 to −6.50 D) in eyes receiving AS drops (Study group) and −2.77 ± 1.64 D (range: −1 to −6 D) in the Control group ($p = 0.145$). The mean follow-up time for the Study and Control groups were 15.2 ± 3.1 months and 15.3 ± 6.1 months, respectively ($p = 0.416$).

All laser ablations were completed successfully without any complications. Mean time for epithelial healing was 2.2 ± 0.25 days in the Study group, and 3 ± 0.0 days in the Control group. Statistically significant ($p = 0.0001$) differences between means of Study and Control groups is 0.8 with a standard deviation of 0.03 and 95 per cent of individual differences ranged from 0.71 to 0.89. All eyes achieved 6/10 or better UCVA at the time of epithelial closure and contact lens removal.

The epithelium healed in two and 2.5 days, respectively, in 60 per cent and 40 per cent of the eyes that received AS drops (Table 1). These patients mostly reported a moderate (Level 2) pain on Day 1, which decreased to mild level (Level 1) afterwards and completely resolved on Day 2 or 2.5 when their contact lenses were removed. Meanwhile, the epithelium healed on Day 3 (100 per cent of the eyes, n = 30) with conventional treatment (Table 1). In four eyes of the Control group, contact lenses were removed on Day 2 with presumed epithelial closure, yet they required replacement of contact lenses due to epithelial defects which healed in Day 3. Patients in the Control group also had moderate pain (Level 2) on Day 1, which decreased to mild (Level 1)

© 2017 Optometry Australia

	Study group	Control group	p-value†
Mean ± standard deviation	2.2 ± 0.25 days	3 ± 0.0 days	0.0001
Minimum–maximum	2–2.5 days	3 days	
Two days (n)	18 eyes (60%)	–	
Two-and-a-half days (n)	12 eyes (40%)	–	
Three days (n)	–	30 eyes (100%)	

n: number of eyes.
†Independent samples t-test.

Table 1. Total duration of epithelial healing

before epithelial closure. However, pain lasted 0.8 days longer in the Control group because of delayed complete epithelial healing.

In the first month, 85 per cent of the eyes in both groups achieved 6/6 or better UCVA. In six months, 100 per cent of eyes achieved 6/7.5 or better UCVA in both groups (Table 2). More than 90 per cent of eyes were within ±0.50 D of emmetropia in six months, which was maintained for 12 months. No eye in either group showed more than +1 haze during the follow-up period (Table 3).

DISCUSSION

Currently, surface ablation procedures are more commonly employed compared to LASIK in order to avoid the complications of LASIK, especially the risk of ectasia.[11] However, the relatively slow visual recovery and early postoperative pain, mostly due to prolonged total duration of epithelial healing, remain the main disadvantages.[12] In our practice, epithelial closure after conventional PRK is completed in three days in most of the cases.[13] Shortening the epithelial healing duration with novel methods may shorten the duration of pain, and decrease other negative aspects of delayed healing, such as risk for haze and infectious keratitis due to a bare epithelial surface. In addition to this, patients may return to their daily routine earlier and the number of postoperative physician visits decreases.

Autologous serum eye-drops are effective in improving epithelial healing in various ocular surface disorders because they positively impact proliferation, migration and differentiation of the eye surface epithelial cells.[14–16] Previous reports of their use after refractive surgery have mostly focused on dry eye. Those studies report a prolongation of the tear break up time and reduction in rose Bengal staining score after LASIK.[17,18] In a recent study, we demonstrated that AS drops can accelerate epithelial healing and shorten postoperative discomfort after LASEK by about one day.[8]

In our practice, the duration of epithelial healing was about a day shorter after PRK compared to LASEK.[13] In this study, in addition to testing the effectiveness of AS eye-drops in epithelial healing after PRK, we intended to develop methods for faster healing following surface ablation. Besides the use of AS drops to accomplish this goal, we have meticulously sized the epithelial opening to 8.0 mm and used fewer eye-drops until epithelial closure. These practices provided a consistent epithelial healing in three days after PRK in the Control group. The healing duration was further shortened by one day in more than half of the eyes in the Study group with the addition of AS drops.

Autologous serum drops were also reported to have a beneficial effect on corneal haze after surface ablation.[19] Similarly, plasma rich in growth factors which act similarly to AS drops was reported to have a positive effect on corneal wound healing in an animal model.[20] The reduction of haze after PRK in the animal model was attributed to faster epithelial healing. In the present study, no eye showed haze intensity greater than +1, and we did not observe a beneficial effect of AS drops on corneal haze. Our study was not designed to provide an evaluation of the effect of AS drops on corneal haze.

Although this was a preliminary study, using AS drops along with meticulous

	Mean ± standard deviation (range)		
	Study group (n = 30)	Control group (n = 30)	p-value†
Preoperative‡	0.95 ± 0.11 (0.8–1.2)	0.96 ± 0.09 (0.8–1.2)	0.898
One month	0.96 ± 0.14 (0.9–1.2)	0.98 ± 0.07 (0.9–1.2)	0.305
Six months	0.97 ± 0.11 (0.8–1.2)	0.99 ± 0.08 (0.8–1.2)	0.430
Last visit§	0.98 ± 0.12 (0.8–1.2)	0.96 ± 0.11 (0.8–1.2)	0.661

†Independent samples t-test.
‡Best spectacle-corrected visual acuity.
§At 12 months or later.

Table 2. Changes in uncorrected Snellen visual acuity over time

	Study group	Control group	p-value
Ablation depth†	66.8 ± 33.8 μm (30–150 μm)	70.4 ± 32.8 μm (24–140 μm)	0.654‡
+1 haze§	Nine eyes	Eight eyes	0.779¶
Preoperative CCT†	551.16 ± 28.7 μm	545.06 ± 31.88 μm	0.439‡

CCT: central corneal thickness.
†Mean ± standard deviation (range).
‡Independent samples t-test.
§At final visit at 12 months or later.
¶Chi-square test.

Table 3. Haze and ablation depth

© 2017 Optometry Australia

epithelial opening were found to be effective in accelerating epithelial healing. Furthermore, use of AS drops caused no adverse effect in our study. The use of AS eye-drops is generally considered safe provided there is strict adherence to preparation and storage protocols.[21,22] An additional topic for future research to hasten epithelial healing after surface ablation may be the investigation of the use of platelet rich plasma, another therapeutic agent similar to AS drops but with higher levels of bioactive mediators. Although its preparation procedure is more sophisticated, positive results with platelet rich plasma after refractive surgery have been reported,[20,23] and it may provide more positive effects compared to AS drops.

In conclusion, use of AS drops accelerate epithelial healing after surface ablation. Further studies with greater patient numbers and longer follow-up would be of interest.

ACKNOWLEDGEMENTS

Ethics committee approval was received for this study from Gazi University Ethics Committee (permit no: 37–2013). No financial support was received for this submission. None of the authors has conflict of interest with this submission.

REFERENCES

1. O'Brart DP. Excimer laser surface ablation: a review of recent literature. *Clin Exp Optom* 2014; 97: 12–17.
2. Ehlke GL, Krueger RR. Laser vision correction in treating myopia. *Asia Pac J Ophthalmol* 2016; 5: 434–437.
3. Santhiago MR, Giacomin NT, Smadja D et al. Ectasia risk factors in refractive surgery. *Clin Ophthalmol* 2016; 10: 713–720.
4. Ambrosio R Jr, Wilson S. LASIK vs LASEK vs PRK: advantages and indications. *Semin Ophthalmol* 2003; 18: 2–10.
5. Azari AA, Rapuano CJ. Autologous serum eye drops for the treatment of ocular surface disease. *Eye Contact Lens* 2015; 41: 133–140.
6. Pan Q, Angelina A, Zambrano A et al. Autologous serum eye drops for dry eye. *Cochrane Database Syst Rev* 2013; 8: CD009327.
7. Gadaria-Rathod N, Lee KI, Asbell PA. Emerging drugs for the treatment of dry eye disease. *Expert Opin Emerg Drugs* 2013; 18: 121–136.
8. Hondur AM, Akcam HT, Karaca EE et al. Autologous serum eye drops accelerate epithelial healing after LASEK. *Curr Eye Res* 2016; 41: 15–19.
9. Tsubota K, Goto E, Fujita H et al. Treatment of dry eye by autologous serum application in Sjögren's syndrome. *Br J Ophthalmol* 1999; 83: 390–395.
10. Fantes FE, Hanna KD, Waring GO 3rd et al. Wound healing after excimer laser keratomileusis (photorefractive keratectomy) in monkeys. *Arch Ophthalmol* 1990; 108: 665–675.
11. Moisseiev E, Sela T, Minkev L et al. Increased preference of surface ablation over laser *in situ* keratomileusis between 2008–2011 is correlated to risk of ectasia. *Clin Ophthalmol* 2013; 7: 93–98.
12. Al-Tobaigy FM. Efficacy, predictability, and safety of laser-assisted subepithelial keratectomy for the treatment of myopia and myopic astigmatism. *Middle East Afr J Ophthalmol* 2012; 19: 304–308.
13. Korkmaz S, Bilgihan K, Sul S et al. A clinical and confocal microscopic comparison of transepithelial PRK and LASEK for myopia. *J Ophthalmol* 2014; 2014: 784185.
14. Kruse FE, Tseng SC. Serum differentially modulates the clonal growth and differentiation of cultured limbal and corneal epithelium. *Invest Ophthalmol Vis Sci* 1993; 34: 2976–2989.
15. Chen YM, Hu FR, Huang JY et al. The effect of topical autologous serum on graft re-epithelialization after penetrating keratoplasty. *Am J Ophthalmol* 2010; 150: 352–359.
16. Lekhanont K, Jongkhajornpong P, Choubtum L et al. Topical 100% serum eye drops for treating corneal epithelial defect after ocular surgery. *Biomed Res Int* 2013; 2013: 521315.
17. Noda-Tsuruya T, Asano-Kato N, Toda I et al. Autologous serum eye drops for dry eye after LASIK. *J Refract Surg* 2006; 22: 61–66.
18. Esquenazi S, He J, Bazan HE et al. Use of autologous serum in corneal epithelial defects post-lamellar surgery. *Cornea* 2005; 24: 992–997.
19. Yee RW, Yee SB. Update on laser subepithelial keratectomy (LASEK). *Curr Opin Ophthalmol* 2004; 15: 333–341.
20. Anitua E, Muruzabal F, Alcalde I et al. Plasma rich in growth factors (PRGF-Endoret) stimulates corneal wound healing and reduces haze formation after PRK surgery. *Exp Eye Res* 2013; 115: 153–161.
21. Hondur A, Bilgihan K, Cirak MY et al. Microbiologic study of soft contact lenses after laser subepithelial keratectomy for myopia. *Eye Contact Lens* 2008; 34: 24–27.
22. Lagnado R, King AJ, Donald F et al. A protocol for low contamination risk of autologous serum drops in the management of ocular surface disorders. *Br J Ophthalmol* 2004; 88: 464–465.
23. Javaloy J, Alió JL, Rodriguez AE et al. Effect of platelet-rich plasma in nerve regeneration after LASIK. *J Refract Surg* 2013; 29: 213–219.

© 2017 Optometry Australia