Changes in ocular surface and Meibomian gland after penetrating Keratoplasty

Kang Yoon Kim1, Byunghoon Chung2, Eung Kweon Kim3,4, Kyoung Yul Seo1, Ikhyun Jun1* and Tae-im Kim1,4*

Abstract

Background: To acquire desirable outcomes of penetrating keratoplasty (PKP), various factors affecting graft survival, visual function, and subjective symptom should be considered. As ocular surface and meibomian gland function are associated with these factors, this study aims to investigate changes of ocular surface and meibomian gland parameters after PKP.

Methods: This retrospective case series study included 24 eyes of 24 patients who underwent penetrating keratoplasty. Examinations on lipid layer thickness (LLT), meiboscore, tear meniscus area (TMA), tear breakup time (TBUT), corneal fluorescein staining (CFS), Schirmer I test (SIT), Ocular Surface Disease Index (OSDI), and meibomian gland functions were performed before and at 1 week, 1 month, 6 months, and 12 months after surgery.

Results: Compared to baseline (2.9 ± 0.6 s), TBUTs were longer at 1 week (4.4 ± 0.5 s, P = 0.027) and 6 months (4.4 ± 0.5, P = 0.048) after surgery. CFS values improved from baseline (6.5 ± 1.1) to 6 months (3.5 ± 0.6, P = 0.023) and 12 months (3.3 ± 0.7, P = 0.001) after surgery. Meibum quality value worsened at 1 week and 12 months after surgery and meibomian gland expressibility value worsened at 1 week and 6 months after surgery compared to baseline. OSDI scores improved at 6 and 12 months after surgery. Meiboscore showed no change throughout the follow up period. The patients with high preoperative meiboscore had worse meibomian gland expressibility at 6 and 12 months and meibum quality at 6 months postoperatively compared to their baseline and to those of patients with low preoperative meiboscore.

Conclusions: After penetrating keratoplasty, ocular surface parameters including corneal staining, TBUT, and OSDI significantly improved whereas meibomian gland parameters showed deteriorations, which was marked in patients with high preoperative meiboscore. Thus, perioperative management of MGD is recommended for patients who undergo penetrating keratoplasty, especially in patients with advanced MGD.

Keywords: Penetrating keratoplasty, Meibomian gland dysfunction, Dry eye disease
Background
Penetrating keratoplasty (PKP) is still a widely performed corneal transplantation procedure for corneal perforation, full thickness corneal opacity, corneal dystrophies, bullous keratopathy, and advanced keratoconus, when the diseased condition is difficult to treat with medications such as ointments, bandage contact lenses, autologous serum, platelet-rich plasma, or lamellar keratoplasty [1–4]. When seeking satisfying outcomes of PKP, it is important to consider various factors that affect graft survival, visual function, and subjective symptoms [1–3]. Common causes for graft failure include allograft rejection, endothelial decompensation, and ocular surface diseases (infectious or sterile keratitis, corneal scarring, etc.) [5]. Additional risk factors for graft failure are preoperative diagnosis, history of ocular infection/inflammation, and ocular surface complications during follow-up (epithelial defect, blepharitis) [6]. Delayed epithelial healing or persistent epithelial defect after keratoplasty, which aggravates infection, melting, scarring, and neovascularization, may restrict graft survival [5, 7]. Even though ocular surface abnormality has been underrated among the factors related to successful clinical outcome after PKP, it not only plays an important role in visual function and subjective symptoms, but is also a vital factor in graft survival [5, 8]. Ocular surface abnormalities with positive corneal staining are common after PKP, and tear film instability has been reported to influence visual function postoperatively [8]. Unstable tear film from the neurotrophic state, aqueous tear deficient state, and persistent ocular surface inflammation after keratoplasty cause dry eye disease (DED), persistent epithelial defect, neovascularization, and even allograft rejection [9].

In keratoplasty, tear film and ocular surface are disrupted by various insults. Keratoplasty severs the corneal nerve and leads to a neurotrophic state, which changes tear secretion, eyelid blinking, and epithelial growth [9, 10]. Changes in ocular surface anatomy affect normal tear film distribution [11]; subsequent tear film instability and dry eye disease in turn damage visual function and ocular surface integrity [12]. In this context, maintaining corneal epithelium, tear film function, and meibomian gland function to achieve homeostasis of the ocular surface is linked to the postoperative outcome of keratoplasty [12, 13].

Previous reports have studied changes of tear function, fluorescein corneal staining, and corneal sensitivity to assess ocular surface abnormalities after keratoplasty [8, 14]. However, despite the ever-growing attention on the role of meibomian gland function in ocular surface homeostasis, ocular surface and meibomian gland dysfunction (MGD) after keratoplasty has not yet been comprehensively studied [13]. In this study, we investigated dry eye disease after keratoplasty by evaluating ocular surface and meibomian gland parameters.

Methods
This retrospective case series study was approved by the Severance Hospital Institutional Review Board, Seoul, South Korea (No. 1–2016-0027) and followed the tenets of the Declaration of Helsinki.

Subjects
Twenty-four eyes from 24 patients who received PKP between January 2017 and December 2018 were enrolled in the study. All the patients were followed up for at least 12 months. Exclusion criteria included a history of previous keratoplasty, limbal deficiency, Sjogren’s syndrome, uncontrolled intraocular pressure, and rejection episode after the surgery. This study included 24 eyes of 24 patients with a mean age of 54.54 ± 14.39 (range, 31–82 years). Indications for keratoplasty were bullous keratopathy (10 eyes), corneal opacity (9 eyes), and advanced keratoconus (5 eyes).

Surgical technique
PKP was performed by a single experienced surgeon (T.I.K.) following standardized procedures under general anesthesia. The recipient cornea trephination diameter was 7 to 8 mm and the donor corneal diameter was the same or 0.25 mm larger than that of the recipient’s. The graft was sutured to the recipient bed with 16 interrupted 10–0 nylon sutures. At the end of the surgery, intraoperative adjustment of sutures was performed to minimize the risk of corneal astigmatism. Pressure patch was applied for 3 to 5 days until epithelial defects are healed.

Postoperatively, topical 0.5% levofloxacin (Cravit, Santen Pharmaceutical, Osaka, Japan) and topical 1% prednisolone acetate (PredForte, Allergan, Irvine, California) were applied four times a day. Topical antibiotics were gradually tapered and maintained for 1 year. Topical steroids were also tapered and maintained for 2 years. Increased intraocular pressure was managed with glaucoma medication and topical 1% prednisolone was switched to topical 0.5% loteprednol (Lotemax, Bausch & Lomb, Rochester, New York) in each patient as needed. Patients were instructed to use 0.1% hyaluronic acid artificial tears for dry eye management, as necessary.

Outcome measures
Detailed examinations of the ocular surface and meibomian gland function were performed by one of the authors (IJ) before surgery and at 1 week, 1 month, 6 months, and 12 months after surgery.
LLT and noninvasive meibography were obtained from LipiView II interferometer (TearScience, Morrisville, NC). Meiboscore was measured from grade 0 (no loss of gland) to grade 3 (loss of more than two-thirds of the total gland area) as previously described [15]. TMA of the lower eyelid was measured using Fourier-domain optical coherence tomography (OCT; RTVue; Optovue, Inc., Fremont, CA). Vertical 2-mm scan images at the middle of the lower eyelid were obtained twice for each eye and TMA was calculated using ReVue software (version 4.0; Optovue, Inc., Fremont, CA) [16]. TBUT was measured three times using a fluorescein strip (Haag-Streit, Koeniz, Switzerland), and the mean value was collected. Subsequent CFS was graded from 0 to 15 according to the National Eye Institute (NEI) scale [17]. SIT without anesthesia was performed for 5 min at least 10 min after corneal staining. Subjective symptoms were assessed using the OSDI questionnaire [18]. Meibomian gland functions were assessed in terms of lid margin abnormality, gland expressibility, and meibum quality. Lid margin abnormalities were graded from 0 to 4 based on the presence of vascular engorgement, plugged Meibomian gland orifices, displacement of the mucocutaneous junction, and irregularity of the lid margin [19]. Meibomian gland expressibility was checked by applying firm digital pressure on the central five glands of the lower eyelid and was measured as grade 0 if all five glands expressed and grade 3 if none of the glands expressed [20]. Meibum quality of eight lower lid glands was graded from 0 (clear) to 3 (toothpaste-like) and a total score of up to 24 was acquired [20].

### Statistical analysis
All data were analyzed using SPSS for Windows version 23.0 (SPSS Inc., Chicago, IL) and expressed as mean ± standard deviation. Repeated measure analysis of variance (RM-ANOVA) was used to analyze the differences between visits. Paired t-test and Wilcoxon signed-rank test were used to compare data between two time points within a group, with adjustment of the level of significance according to the Bonferroni correction. Subgroup analysis and comparison with different characteristics were performed using independent samples t-test and Mann-Whitney test. P values less than 0.05 were considered statistically significant.

### Results

**Ocular surface and Meibomian gland status after Keratoplasty**

The ocular surface and meibomian gland parameters of DED after keratoplasty are shown in Table 1. TBUT values were higher at 1 week and 6 months after surgery than the baseline. Corneal staining scores showed a significant reduction from the baseline at 6 and 12 months after surgery. Meibum quality values were elevated at 1 week and 12 months after surgery. Meibomian gland expressibility values were elevated at 1 week and 6 months after surgery. Meiboscore showed no change throughout the follow-up period. LLT was higher at 1 week postoperatively than the baseline. OSDI scores were improved at 6 and 12 months after surgery compared to the baseline. Other clinical parameters of DED after surgery were not significantly different from the

| Clinical Parameters of Dry Eye Disease | Baseline | 1 week | 1 month | 6 months | 12 months |
|---------------------------------------|----------|--------|---------|----------|----------|
| Tear meniscus area (10^9 mm^2)        | 40 ± 8   | 52 ± 9 | 42 ± 10 | 29 ± 7   | 54 ± 6   |
| TIBUT (sec)                           | 2.9 ± 0.6| 4.4 ± 0.5 | 3.9 ± 0.5 | 4.4 ± 0.5 | 4.3 ± 0.7 |
| Corneal fluorescein staining score (0–15) | 6.5 ± 1.1 | 6.7 ± 0.8 | 5.1 ± 0.7 | 3.5 ± 0.6 | 3.3 ± 0.7 |
| Schirmer's test I value (mm/5')       | 15.4 ± 2.3| 17.8 ± 2.2 | 13.4 ± 2.4 | 16.2 ± 2.3 | 16.5 ± 2.2 |
| Lid margin abnormality (0–4)         | 1.9 ± 0.3| 2.1 ± 0.2 | 1.9 ± 0.2 | 1.9 ± 0.2 | 2.0 ± 0.3 |
| Meibum quality (0–24)                | 8.8 ± 1.4| 11.3 ± 1.1 | 10.7 ± 1.2 | 11.0 ± 1.3 | 12.7 ± 1.1 |
| Meibomian gland expressibility (0–3)  | 1.3 ± 0.2| 1.8 ± 0.2 | 1.6 ± 0.2 | 2.0 ± 0.2 | 1.7 ± 0.2 |
| Meiboscore (0–3)                      | 1.2 ± 0.2| 1.3 ± 0.2 | 1.2 ± 0.2 | 1.3 ± 0.2 | 1.3 ± 0.2 |
| Lipid layer thickness (nm)            | 79.0 ± 4.1| 95.6 ± 2.5 | 86.9 ± 3.6 | 86.7 ± 3.8 | 81.1 ± 4.1 |
| OSDI (0–100)                          | 40.4 ± 4.2| 39.7 ± 4.28 | 34.13 ± 4.42 | 23.14 ± 3.52 | 28.78 ± 3.37 |

*TBUT tear break up time, OSDI ocular surface disease index
Results are expressed as mean ± SD
*Statistical significance (P < 0.05)
Changes in parameters in groups with different Meiboscores

Meiboscore of each patient remained unchanged from the baseline through the whole follow-up period. Patients were divided into two groups with different meiboscores; group 1, levels 0–1 and group 2, levels 2–3 from the baseline examination. Ocular surface parameters and meibomian gland parameters between the two groups at each time point were compared (Fig. 2). Compared to the baseline, the high meiboscore group showed significant worsening of meibomian gland expressibility values at 6 and 12 months and meibum quality values at 6 months after surgery. When compared to the low meiboscore group, meibomian gland expressibility values in the high meiboscore group were significantly worse at 6 and 12 months after surgery, and meibum quality values in the high meiboscore group were significantly worse at 6 months after surgery (Table 2). Other parameters showed no statistical differences between the groups at each time point.

Effect of Antiglaucoma medication usage on parameters

The number of patients who used antiglaucoma medications for over 1 month was 11 and those who did not were 13. These two groups of patients were compared to analyze the effect of antiglaucoma medications on the entire ocular surface and meibomian gland parameters. SIT values were significantly higher in the group that used antiglaucoma medications in the pre- and postoperative period.

Discussion

In this study, we analyzed changes in the ocular surface and meibomian gland parameters over 12 months of follow-up for PKP. We found that the corneal fluorescein score, TBUT, and OSDI showed a significant improvement over time. We also found that meibomian gland functions, such as meibomian gland expressibility and meibum quality, significantly deteriorated without structural changes after PKP, and that the extent of deterioration was more prominent in patients with preexisting MGD.

PKP has been a widely used corneal transplantation procedure for various corneal disorders. This procedure...
**Fig. 2** Clinical parameters of dry eye disease in groups with different meiboscores. Group 1, meiboscore 0–1; Group 2, meiboscore 2–3.

*Statistical significance ($P < 0.05$) for changes between groups at each time point

|                      | Group 1 | $P$ (vs Baseline) | Group 2 | $P$ (vs Baseline) | $P$   |
|----------------------|---------|-------------------|---------|-------------------|-------|
| **Meibomian gland expressibility (0–3)** |         |                   |         |                   |       |
| Baseline             | 1.2 ± 0.9 |                   | 1.7 ± 0.8 |                   | 0.198 |
| 1 week               | 1.7 ± 0.9 | 0.046$^a$         | 2.0 ± 1.0 | 0.356             | 0.494 |
| 1 month              | 1.7 ± 1.0 | 0.095             | 1.4 ± 0.8 | 0.356             | 0.536 |
| 6 months             | 1.7 ± 0.9 | 0.149             | 2.7 ± 0.5 | 0.038$^a$         | 0.006$^b$ |
| 12 months            | 1.4 ± 1.0 | 0.455             | 2.6 ± 1.1 | 0.048$^a$         | 0.02$^b$ |
| **Meibum quality (0–24)** |         |                   |         |                   |       |
| Baseline             | 7.9 ± 7.2 |                   | 11.0 ± 6.2 |                   | 0.325 |
| 1 week               | 11.5 ± 6.0 | 0.021$^a$         | 10.9 ± 4.8 | 0.915             | 0.796 |
| 1 month              | 11.5 ± 6.0 | 0.034$^a$         | 9.7 ± 5.3 | 0.580             | 0.611 |
| 6 months             | 7.9 ± 4.1 | 0.970             | 18.4 ± 5.0 | 0.045$^a$         | 0.001$^b$ |
| 12 months            | 11.4 ± 4.7 | 0.086             | 15.7 ± 5.8 | 0.168             | 0.07  |

Group 1, meiboscore 0–1; Group 2, meiboscore 2–3

Results are expressed as mean ± SD

$^a$Statistical significance ($P < 0.05$) between baseline and follow up period in each group

$^b$Statistical significance ($P < 0.05$) between two groups at each time point
involves removal of the full-thickness cornea, total sev-
erance of the corneal nerve, intraocular manipulation, and
extensive suturing with resulting ocular surface irregu-
larity. Even after uneventful surgery, ocular discomfort
occurs. A previous study reported that superficial puncta-
tate keratopathy and dry eye were common complica-
tions of corneal denervation after PKP [21]. Aggravation
of DED parameters, such as TBUT, corneal fluorescein
stain, Schirmer I test, and corneal esthesiometry, after
keratoplasty, has been previously reported [11, 14, 22].
Distinct from previous reports, the current study investi-
gated meibomian gland parameters in addition to ocular
surface parameters as the two are not separable.

Corneal staining scores showed gradual improvement
from 1 week after surgery to 12 months after surgery.
However, some degree of epithelial damage persisted
even after 12 months. The poor corneal staining scores
at the baseline were caused by primary corneal diseases.
As the preoperative diagnosis of cornea was treated by
keratoplasty and harmful injuries on epithelium induced
by pathologic corneas were ceased, the corneal epithe-
lium regained its integrity and staining scores improved
from baseline which is consistent with a previous report
by Lin et al. [14] Corneal epithelium defect after kerato-
plasty gradually improved but did not completely heal
until the last study visit. The remaining epithelial defect
after keratoplasty are attributable to denervation, discon-
nection from limbal stem cells, frequent use of eye drops,
and aggravation of DED [9, 22]. And deterioration of
meibomian gland function which was observed in our
study also causes epithelial damage via released inflam-
atory mediators and lipids on the ocular surface [23].

Compared to the measurement at the baseline, TBUTs
significantly improved at 1 week and 12 months after
surgery. Despite the fact that tear meniscus areas and
Schirmer I test values showed no significant changes
compared to those of baseline, the increase in TBUTs
demonstrates an improvement in tear film stability with-
out changes in tear production or volume. OSDI scores
at 6 and 12 months after surgery were lower than those
at the baseline. Surgical removal of pathologic cornea
and subsequent corneal denervation after keratoplasty
may have mitigated dry eye sensation at the early post-
operative period. However, restoration of corneal sensa-
tion and deterioration of MGD led to increase in OSDI
scores at 12 months after surgery.

Comprehensive assessment of meibomian gland pa-
rameters using slit-lamp microscopy, meibography, and
interferometry was performed. Meibomian gland expres-
sibility worsened at 1 week and 6 months after surgery.
Meibum quality scores showed a trend of worsening
after surgery, and the changes, compared to those at
the baseline, were significant at 1 week and 12 months
postoperatively. Lid margin abnormality score and
meiboscore showed no statistically significant change
during the follow-up period. MGD is caused by the stag-
nation of meibum inside the glands, dilation of the
ductal system, and consequent loss of glands. Our hy-
pothesis was that PKP may aggravate all aspects of
MGD. However, only the functional parameters of mei-
bomian glands, such as expressibility and meibum qual-
ity, were altered without any structural changes in the
lid margin and meibomian gland tissues.

Causes of functional changes in meibomian glands are
multifactorial, and the exact underlying mechanism is
unclear. Damage in neural regulation of meibomian
glands may affect meibum secretion. Meibomian glands,
which are surrounded by a dense meshwork of choliner-
gic parasympathetic nerve fibers may have a role in the
neural feedback loop [24, 25]. As the denervation of
the cornea by keratoplasty is known to interfere with the
feedback loop of the lacrimal functional unit [23], a pos-
sible neural dysregulation of meibomian glands after
keratoplasty may contribute to meibum secretion
abnormalities.

Insufficient lid hygiene after keratoplasty may contrib-
ute to the functional deterioration of the meibomian
glands. Lid hygiene is known to reduce lipid by-products
and lipolytic bacteria on the lid margin, which can re-
duce the level of ocular surface MMP-9, improve the
quality of the lipid layer, and alleviate MGD [26–28].
After keratoplasty, patients are usually discouraged from
cleaning their eyelids because of the risk of mechanical
damage to the ocular surface. Lack of lid hygiene, which
causes stagnation of lipids and obstruction of the gland,
may cause functional changes of meibomian gland after
surgery.

The effects of preservatives and antiglaucoma medica-
tions on meibomian glands have been previously re-
ported, and are known to cause functional and
structural changes in meibomian glands [29–31]. Anti-
glaucoma medications cause subclinical inflammation
which results in keratinization of meibomian gland ori-
fice and subsequent stagnation of meibum [29]. Lee
et al. demonstrated worsening of lid margin abnormality,
expressibility of meibum, and meiboscore in patients
using preservative-containing antiglaucoma medications
[30]. However, in our study, the groups with or without
antiglaucoma medications only differed in SIT values.
Because our study was unable to control the variables
related to antiglaucoma medication usage, the results
could not reflect the impact of antiglaucoma medication
on the ocular surface and meibomian glands. Taken to-
going, the mechanism of MGD after keratoplasty may
involve various insults, including damaged neural regula-
tion of meibomian glands, insufficient lid hygiene,
preservative-containing eyedrops, and antiglaucoma
medications.
Functional changes of meibomian glands after surgery have been previously reported in surgeries other than PKP [32–36]. Unavoidable trauma from cataract surgery may induce ocular inflammation [37]. Toxicity from eye drops [38] and lid dysfunction because of intraoperative use of lid speculum may also contribute to MGD. Our results are similar to those reported for cataract surgeries in that no structural changes occurred while meibomian gland function deteriorated. It can be speculated that cataract surgery interferes with the ocular surface and meibomian glands in ways similar to PKP, but their exact mechanism could not be elucidated in the current study.

At the baseline, patients were divided into two groups; high and low meiboscore groups, and the parameters between the two groups were compared. Functional parameters, meibomian gland expressibility, and meibum quality score showed statistically significant differences between the two groups. High meiboscore group had worse expressibility and meibum quality at 6 and 12 months after surgery. Patients with worse preexisting MGD were more prone to the functional deterioration of meibomian glands without any change in meiboscore. MGD starts with the stagnation of meibomian gland secretion, and chronic inflammation within the glands leads to subsequent structural changes. Therefore, patients with structural damage of the glands are more likely to have long-standing MGD and frail gland function, which could easily be affected by harmful insults that lead to functional degradation of the glands.

As a limitation of our study, the sample size was relatively small due to infrequent cases of keratoplasty. Moreover, a control group was lacking. In addition, there could be numerous variables affecting the DED parameters that were not addressed in our study, which can confound the results. Furthermore, different use of eye drops, such as steroids, antiglaucoma medications, and artificial tears, among study participants could not be controlled and could have affected the results. Increased intraocular pressure after keratoplasty is unpredictable and the response to treatment is not uniform. Thus, use of antiglaucoma medication after surgery is individualized. The number of antiglaucoma medications and duration of use are too heterogeneous in patients and very complicated to control. Further studies with meticulous study design are needed to assess the effect of the antiglaucoma medication in post-keratoplasty patients.

In spite of these limitations, our study showed significant clinical improvement in ocular surface conditions, including better corneal staining and OSDI, and elongation of TBUT. Although greater deterioration of the functional parameters of MGD after PKP was observed for the entire observation period, especially among the patients with an advanced stage of MGD, our findings suggest that thorough observation of MGD before and after keratoplasty is necessary to identify and manage ocular surface and meibomian gland deficits to achieve more desirable post-keratoplasty results.

**Abbreviations**

PKP: Penetrating keratoplasty; LLT: Lipid layer thickness; TMA: Tear meniscus area; TBUT: Tear breakup time; CFS: Corneal fluorescein staining; ST: Schirmer I test; OSDI: Ocular Surface Disease Index; DED: Dry eye disease; MGD: Meibomian gland dysfunction

**Acknowledgements**

Not applicable.

**Authors’ contributions**

KYK, IJ, and TIK designed the study; KYK, BC, and IJ assisted with data acquisition and interpretation; KYK, BC, and IJ performed statistical analyses; KYK, BC, ERK, KY, IJ, and TIK contributed to the discussion; KYK and TIK drafted the manuscript; and IJ and TIK revised the manuscript. All authors read and approved the final manuscript.

**Funding**

This research was supported by Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Education, Science and Technology (NRF-2019R1F1A1062468).

**Availability of data and materials**

The datasets generated analyzed during the current study are not publicly available due to patient’s data privacy but can be made available from the corresponding author upon reasonable request.

**Ethics approval and consent to participate**

This study was approved by the Institutional Review Board of Severance Hospital (IRB No 1–2016-0027) by which the requirement for informed consent was waived because of the retrospective nature of this study.

**Consent for publication**

Not applicable.

**Competing interests**

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as potential conflicts of interest.

**Author details**

1. Department of Ophthalmology, Institute of Vision Research, Yonsei University College of Medicine, Seoul, Republic of Korea. 2. Department of Ophthalmology, International St. Mary’s Hospital, Catholic Kwandong University College of Medicine, Incheon, Republic of Korea. 3. Saevit Eye Hospital, Goyang, Republic of Korea. 4. Corneal Dystrophy Research Institute, Department of Ophthalmology, Yonsei University, College of Medicine, Seoul, Republic of Korea.

**Received:** 10 December 2020 **Accepted:** 27 January 2021

**Published online:** 15 February 2021

**References**

1. Ing JJ, Ing HH, Nelson LR, Hodge DO, Bourne WM. Ten-year postoperative results of penetrating keratoplasty. Ophthalmology. 1998;105(10):1855–65.
2. Muraine M, Sanchez C, Watt L, Retout A, Brasseur G. Long-term results of penetrating keratoplasty. A 10-year-plus retrospective study. Graefes Arch Clin Exp Ophthalmol. 2005;241(7):571–6.
3. Ono T, Ishiyama S, Hayashida T, Mori Y, Nejima R, Miyata K, Amano S. Twelve-year follow-up of penetrating keratoplasty. Jpn J Ophthalmol. 2017;61(2):131–6.
4. Rechichi M, Ferrise M, Romano F, Gallelli L, Toschi V, Dominjanni A, Meduri A. Autologous platelet-rich plasma in the treatment of refractory corneal ulcers: a case report. Am J Ophthalmol Case Rep. 2020;20:100838.
5. Price FW Jr, Whitson WE, Collins KS, Marks RG. Five-year corneal graft survival. A large, single-center patient cohort. Arch Ophthalmol. 1993;111(6):799–805.

6. Fasolo A, Capuzzo C, Forme A, Franch A, Birattari F, Canto G, Cucco F, Prosodcino G, Sala M, Delle Noci N, et al. Risk factors for graft failure after penetrating keratoplasty: 5-year follow-up from the corneal transplant epidemiological study. Cornea. 2011;30(12):1328–35.

7. Constantinou M, Jhanji V, Wu JM, Srikumaran S, Amano S. Effects of long-term topical anti-glaucoma medications on meibomian glands. Graefes Arch Clin Exp Ophthalmol. 2012;250(8):1144–50.

8. Sama A, Jhanji V, Wu JM, Srikumaran S, Amano S. Effects of long-term topical anti-glaucoma medications on meibomian glands. Graefes Arch Clin Exp Ophthalmol. 2012;250(8):1144–50.

9. Shriver J, Jhanji V, Wu JM, Srikumaran S, Amano S. Effects of long-term topical anti-glaucoma medications on meibomian glands. Graefes Arch Clin Exp Ophthalmol. 2012;250(8):1144–50.

10. Muller LJ, Marfurt CF, Kruse F, Tervo TM. Corneal nerves: structure, contents and function. Exp Eye Res. 2003/06(5):521–42.

11. Hiraizumi M, Satake Y, Hirayama M, Shimazaki-Den S, Konomi K, Shimazaki J. Changes in corneal sensation, epithelial damage, and tear function after descemetic stripping automated endothelial keratoplasty. Cornea. 2013;32(9):1255–9.

12. Craig JP, Nelson JD, Azar DT, Belmonte C, Bron AJ, Chauhan SK, de Paiva CS, Gomes JAP, Harnett MM, Jones L, et al. TFOS DEWS II report executive summary. Ocul Surf. 2017;15(4):802–12.

13. Nichols KK, Foukis GN, Bron AJ, Glasgow BJ, Dogru M, Tsirouka R, Lemp MA, Sullivan DA. The international workshop on meibomian gland dysfunction: executive summary. Invest Ophthalmol Vis Sci. 2011;52(4):1292–2.

14. Lin X, Xu B, Sun Y, Zhong J, Huang W, Yuan J. Comparison of deep anterior lamellar keratoplasty and penetrating keratoplasty with respect to postoperative corneal sensitivity and tear film function. Graefes Arch Clin Exp Ophthalmol. 2014;252(11):1779–87.

15. Arita R, Itoh K, Inoue K, Amano S. Noncontact infrared meibography to document age-related changes of the meibomian glands in a normal population. Ophthalmology. 2008;115(9):1911–5.

16. Nguyen P, Huang D, Li Y, Sidda SR, Ramos S, Pappuru RR, Yu SC. Correlation between optical coherence tomography-derived assessments of lower tear meniscus parameters and clinical features of dry eye disease. Cornea. 2012;31(6):680–5.

17. Lemp MA. Report of the National eye Institute/industry workshop on clinical trials in dry eyes. CLAO J. 1995;21(4):221–32.

18. Schiffman RM, Christianson MD, Jacobsen G, Hirsch JD, Reis BL. Reliability and validity of the ocular surface disease index. Arch Ophthalmol. 2000;118(6):615–21.

19. Jung JW, Park SY, Kim JS, Kim EK, Seo KY, Lee H, Min K, Kim TI. Minocycline controls clinical outcomes and inflammatory cytokines in moderate and severe meibomian gland dysfunction. Am J Ophthalmol. 2012;154(6):949–58.

20. Lee H, Min K, Kim TI. Minocycline controls clinical outcomes and inflammatory cytokines in moderate and severe meibomian gland dysfunction. Am J Ophthalmol. 2012;154(6):949–58.

21. Sama A, Jhanji V, Wu JM, Srikumaran S, Amano S. Effects of long-term topical anti-glaucoma medications on meibomian glands. Graefes Arch Clin Exp Ophthalmol. 2012;250(8):1144–50.

22. Lee SY, Lee K, Park CK, Kim S, Bae HW, Seong GJ, Kim CY. Meibomian gland dropout rate as a method to assess meibomian gland morphologic changes during use of preservative-containing or preservative-free topical prostaglandin analogues. PLoS One. 2019;14(6):e0218886.

Publisher’s Note

Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.