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Original Article

COMBINED CRYOPRESERVED AMNIOTIC MEMBRANE, AUTOLOGOUS SERUM EYE DROPS, AND TEA-TREE OIL LID SCRUB THERAPY FOR THE TREATMENT OF KERATOCONJUNCTIVITIS IN A *STAPHYLOCOCCUS* HYPERSENSITIVE TEENAGER

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ABSTRACT

Background and Objectives

The purpose is to present an interventional case report detailing the novel treatment of recurrent bilateral keratoconjunctivitis secondary to *Staphylococcus* hypersensitivity in a teenage female. The patient's disease course was marked by recurrent episodes of severe bilateral catarrhal infiltrative or phlyctenular keratoconjunctivitis over a period of five years. Previous comprehensive corneal and conjunctival cultures had been performed twiceyielding only growth of *Staphylococcus aureus* and *epidermidis*. Prior short-term treatment regimens of topical steroids and antibiotics had not prevented recurrences.

Methods

This is an interventional case reportcomparing therapeutic placement of a self-retained cryopreserved amniotic membrane, Prokera ® Slim (PKS by BioTissue), after one week of topical steroid treatment with difluroprednate 0.05% qid in the right eye (OD) to a steroid (difluroprednate 0.05%) taper in the left eye (OS) over an approximate one-month time period. Subsequent long-term maintenance therapy included autologous serum eye drops (ASED) dosed six times daily and tea tree oil containing lids scrubs (SteriLid Foam ® by TheraTears) two times daily, until the present time. Supportive meibomian gland expression for meibomian gland dysfunction was also performed at the three-month treatment mark.

Results

The PKS treatment in the right eye resulted in superior visual acuity and ocular comfort when compared to a steroid taper in the left eye over an approximate one-month time period. These results were maintained in the right eye at the 12-month time frame. The right eye experienced a recurrence when the patient self-discontinued all therapy in both eyes at the four-month mark. With re-initiation of maintenance therapy of ASED and tea tree oil containing lids scrubs, she has remained symptom free for over eight months in the right eye.

Conclusions

Further comparison and study of the treatment modalities above could result in a potentially faster, safer, and superior treatment and prevention protocol for topical ocular inflammation in *Staphylococcus* hypersensitive keratoconjunctivitis, particularly in pediatric populations.

Staphylococcus aureus and *epidermis* bacterial species are commonly found on the eyelid and ocular surface. In low quantities, these bacteria generally do not cause opportunistic infection or inflammation.[1–3] However, in patients with *Staphylococcus* hypersensitivity these bacteria can lead to an inflammatory keratoconjunctivitis, as a result of a delayed type IV hypersensitivity reaction.[4–6] Hallmarks of this type of antigen-antibody, immune complement activation can include catarrhal infiltrates (generally at the two-, four-, eight-, and ten- o clock positions and separated from the limbus by a few millimeters), phlyctenules, punctate epithelial erosions at the lower one third of the cornea, and corneal ulcers.[5–7,9] This case details novel treatment and prevention protocols in a patient with *Staphylococcus* hypersensitivity.

CASE REPORT

Presentation

A 19-year-old Hispanic female presented for emergency evaluation to a primary care setting. The chief complaint reported was red, irritated, eyes, as well as constant blur and photophobia for two-days. At least three times yearly, for five years, she had experienced episodes of severe bilateral, catarrhal infiltrative, and phlyctenular keratoconjunctivitis. The patient desired alternative therapy to stop the recurrent episodes. The patient reported make-up and contact lens intolerance. Quality of life issues were a concern as she reported frequent doctor visits were interfering with her ability to function at work and school. She noted that vision and comfort out of her right eye had been worse since the series of red eyes began when she was 14-years old. She reported mild, chronic irritation in-between flare-ups.

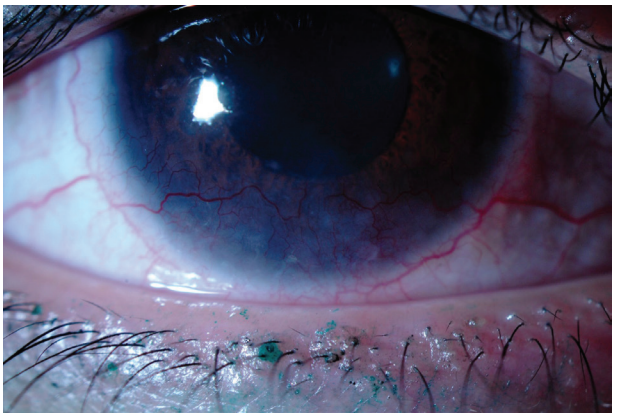
Corneal and conjunctival cultures had been performed twice, with *Staphylococcus aureus* and *epidermidis* isolated from the corneal scrapings and conjunctival swabs. Polymerase chain reaction showed negative results for herpes simplex virus, varicella zoster virus, and cytomegalovirus. No fungal or other bacterial species was isolated. *Acanthamoeba polyphaga* was ruled out. Prior treatment regimens of topical ocular steroids and antibiotics provided temporary relief for no longer than three to four months at a time. She confirmed a history of multiple chalazia or hordeola throughout the past five years which always subsided with hot compresses. The patient denied frequent colds, upper respiratory infections, allergies, or asthma. She denied any serological or blood testing performed for this condition. The patient was taking an estrogen-based oral contraceptive but denied exposure to any sexually transmitted disease.

Visual acuity was reduced to 20/60 OD and 20/30 OS with no improvement with pinhole. Keratoconjunctivitis was confirmed with slit lamp examination (Figure 1A, 1B). Both eyes manifested diffuse corneal edema and punctate erosions denser at the lid seal position. There were multiple corneal infiltrates, all at least 2 mm from the limbus. Neovascularization of the inferior cornea, extending two mm below the pupillary margin was visualized on both eyes, and trace neovascularization at the edge of the limbus was present in all other meridians (Figure 1A). There was no significant anterior chamber reaction. Grade 1 cylindrical dandruff was detected at the lash bases on all four eyelids. There was no significant papillary or follicular response seen on the palpebral conjunctiva. Diffuse grade 2+ injection of the bulbar conjunctiva was evident.

Staphylococcus hypersensitivity was the presumed primary culprit for the inflammation manifested, and the patient was placed on a topical ocular steroid, Durezol (difluprednate ophthalmic emulsion 0.05%), four times daily (qid) for one week, along with twice daily (bid) lid scrubs containing tea-tree oil (SteriLid Foam ® by TheraTears). The patient was asked to return in one week to determine the efficacy of the treatment regimen.

At the one-week treatment follow-up there was mild improvement in visual acuity to 20/40 OD and 20/25 OS. The eyelids were clean of any cylindrical dandruff and the conjunctival injection had calmed to grade 1+. Corneal inflammation was improving, but moderate keratitis, corneal edema and resolving infiltrates remained in both eyes. Meibography confirmed the patient had grade 1 meibomian gland loss seen as truncation of the lower eyelid meibomian glands (Figure 2). Her non-invasive tear break-up time (as measured with the Oculus Keratograph ® 5M), was reduced at 2.49 seconds OD and 4.59 seconds

FIG. 1 Keratoconjunctivitis secondary to Staphylococcus hypersensitivity at presenting visit and 12 month following initial presentation (a) right eye ocular surface initial presentation, (b) left eye ocular surface initial presentation, (c) right eye punctate keratitis at 12 months, (d) right eye ocular surface at 12 months, (e) left eye keratitis at 12 months, (f) left eye ocular surface at 12 months.



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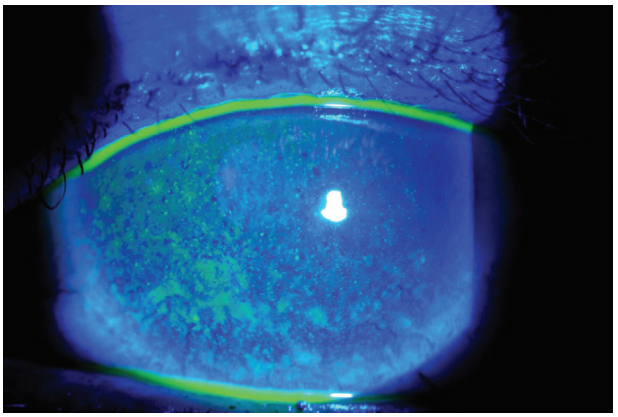

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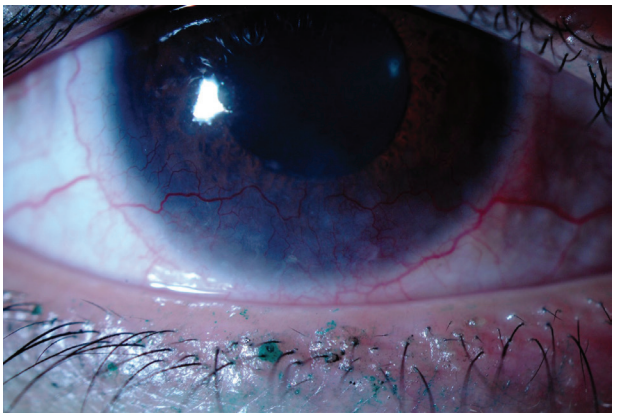

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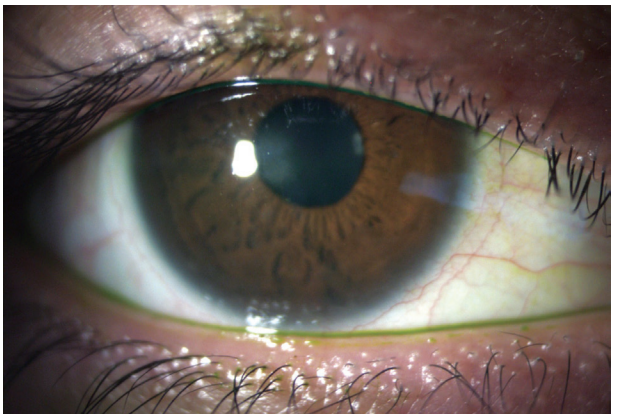

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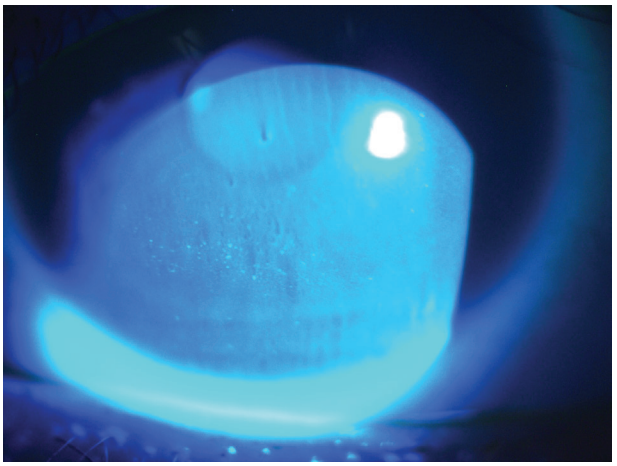

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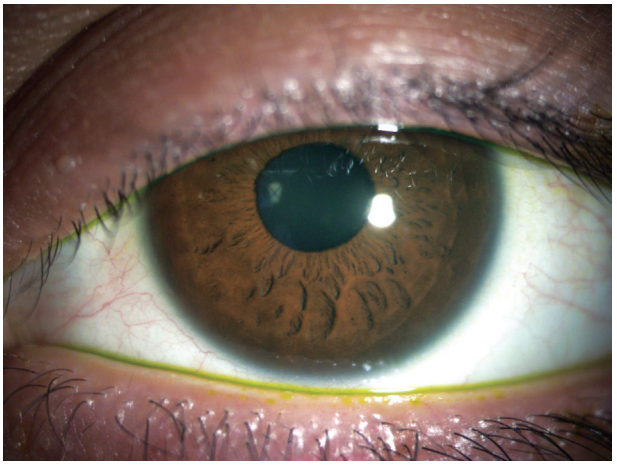

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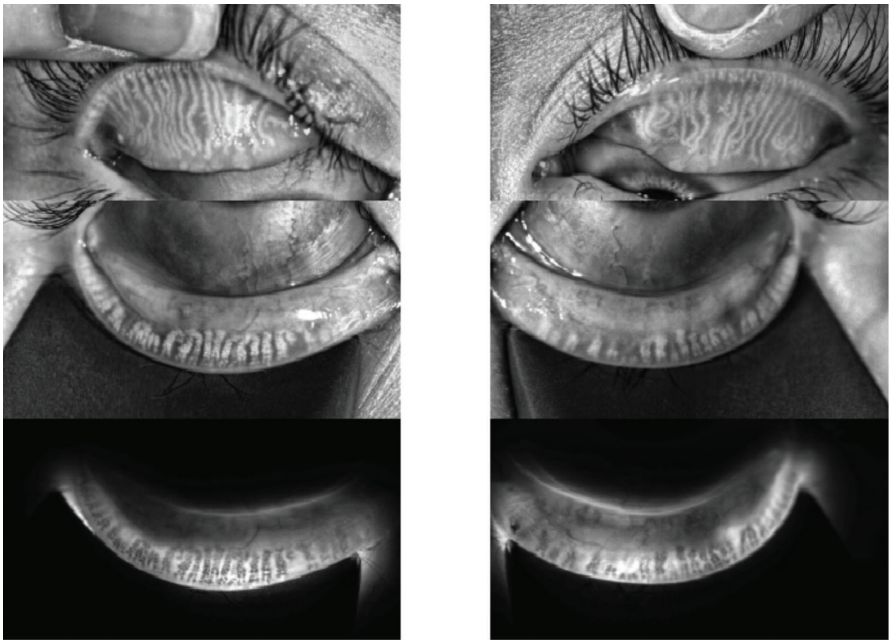
FIG. 2 Meibography imaging showing meibomian gland truncation in lower eyelids.



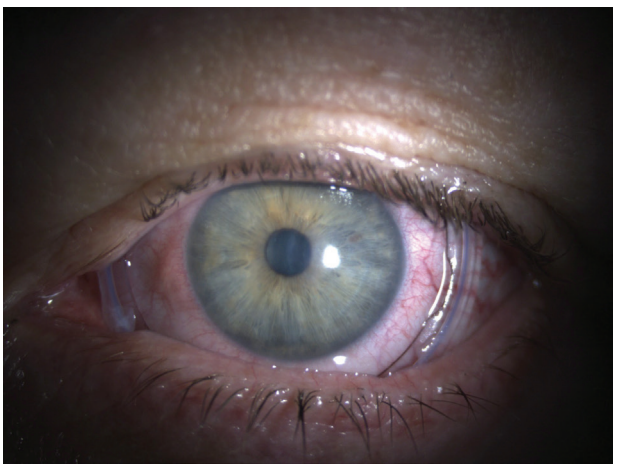
OS. The lipid layer of the tear film, via white light interferometry (with LipiView II by Tears Science), was reduced to borderline levels of 63 nm OD and 60 nm OS. Less than 60 nm of lipid layer thickness is correlated with dryness symptoms.[10]

A Prokera Slim cryopreserved amniotic membrane (PKS) was placed on the ocular surface of the right eye, with all other treatments (lid scrubs and steroids) to be discontinued. An example image of a PKS cryopreserved amniotic membrane on the ocular surface can be seen in Figure 3. The left eye continued to receive Durezol, tapered to three times daily, along with tea tree oil containing lid scrubs twice daily.

After five days of PKS treatment, the membrane ring in the right eye was removed. Durezol drops were tapered to twice daily in the left eye. Tea tree oil containing lid scrubs were to be continued twice per day in both eyes

The patient returned once weekly for two more weeks, tapering the Durezol at each visit by one drop daily in the left eye. The tea tree oil containing lid scrubs were continued twice daily in both eyes.

FIG. 3 Example image of Prokera Slim cryopreserved amniotic membrane on ocular surface.

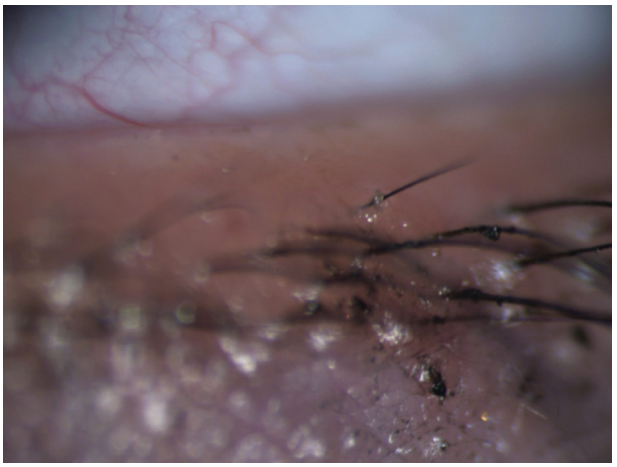


At the 3.5-week mark, visual acuity was measured at 20/25 OD and OS. The patient subjectively reported that the vision in the right eye was clearer than left eye, which had not been the case for five years. She

also reported the right eye was far more comfortable, with a sensation of "smoothness" in blinking. The punctate staining was reduced to grade 1+ inferiorly in both eyes, with less density OD compared to OS. Durezol was discontinued. Autologous Serum Eye Drops (ASED) were prescribed six-times daily, along with twice daily tea tree oil containing lid scrubs for continued maintenance. The patient was approved to return to primary eye care for her annual refraction, which resulted in best corrected visual acuity of 20/15 OD and 20/20 OS at the six-week mark.

At 12 weeks from the initial appointment, the patient was asymptomatic as long as she continued daily intervention. She had self-tapered her lid scrubs to once daily, before bed and only used ASED two to four times daily. Visual acuity through her new glasses was 20/20+2 OD and 20/20 OS. A few rare cylindrical dandruff-coated eyelashes at the lash base were noted on slit lamp examination (Figure 4), with trace inferior corneal punctate epithelial erosions at the lid-seal location in both eyes. Her non-invasive TBUT was mildly improved, but still reduced to 3.06 seconds OD and 5.35 seconds OS. The lipid layer thickness was still reduced to borderline findings of 52 nm OD and 45 nm OS. Her meibomian gland secretion scores were also borderline at six clear secreting glands OD and OS. Eyelid margins were debrided with a golf spud, followed by a 10-minute micro-bead hot compress application and subsequent meibomian gland expression (with a combination of a Mastrota paddle and Arita forceps (Figure 5A and 5B). Tea tree oil containing lid scrubs twice daily and ASED four times per day were recommended.

FIG. 4 Cylindrical dandruff at eyelash base.



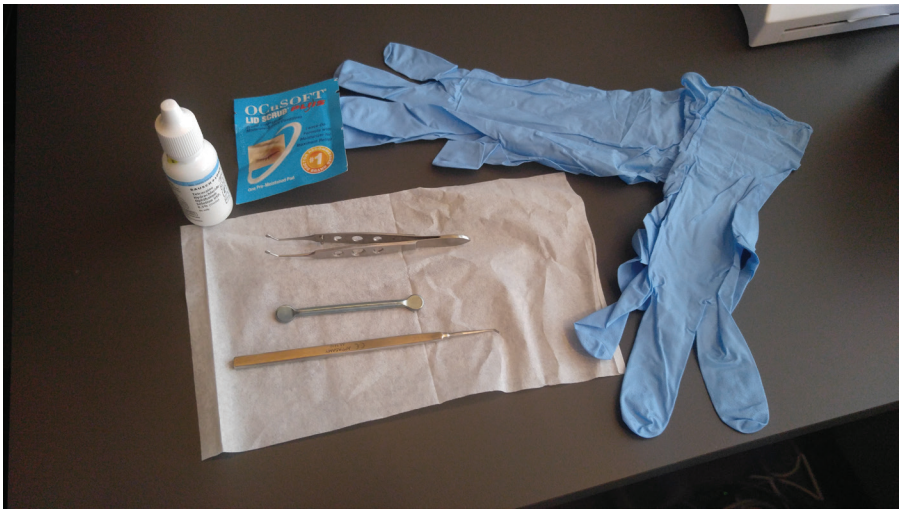
At month four, the patient once again presented in a primary care clinic with a recurrence of phylectenular keratitis OD. She reported that after the 3-month follow-up she had felt asymptomatic so she had discontinued all therapy. Repeat PKS treatment was cost-prohibitive for this non-medically insured patient, so she elected for a one month long steroid taper regimen with Durezol in tha right eye. The patient reported full understanding of the underlying cause of her recurrence and agreed to return to her regimen of tea tree oil containing lid scrubs twice daily and ASED four times daily.

With strict daily adherence to tea tree oil containing lid scrubs and ASED, the patient has experienced the longest time interval between occurrences. She was recurrence free for nearly one year in the left eye and 8 months in the right eye. Her visual acuity remained steady at 20/20 OD and OS one year after the initial presentation. While keratitis was still present, it is very mild and located only at the lid seal position (see Figure 1C and 1E). Table 1 shows the full treatment protocol over the one-year time period.

Clinical Manifestations/Evaluation

Historical eyelid disease in combination with signs of inflammation at this presentation are crucial in determining the correct diagnosis. Past eyelid disease (e.g., chalazia/hordeola) and phlyctenules have been well documented in the patient's chart. In addition, this presentation showed corneal infiltrates at least two mm from the limbus and blepharitis. Inflammation of the corneal and conjunctiva were also consistent with Bierdman, et al.,[11,12] whom identified the presence of gamma exotoxins released by *Staphylococcal* species as potentially causative for corneal and conjunctival inflammation. The recurrent nature of these symptoms and the *Staphylococcus* species identified with ocular culturing in the past, suggest a *Staphylococcus* hypersensitive keratoconjunctivitis.[4–11] There was no anterior chamber reaction, nor any significant papillary or follicular response, which is consistent with *Staphylococcus* hypersensitivity. She had experienced

FIG. 5 (A) Equipment for hand meibomian gland expression; (B) Hot compress application prior to meibomian gland expression.



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improvement, though temporary, with topical ocular steroids and antibiotics, which are the classic treatments for this condition.[4–13]

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TABLE 1 Treatment Timeline for *Staphylococcus* Hypersensitive Patient

Timeline	Treatment OD	Treatment OS
Initial Presentation	• Durezol bid • Thera Tears SteriLid Scrub bid	• Durezol bid • Thera Tears SteriLid Scrub bid
One Week	• Prokera Slim cryopreserved amniotic membrane (PKS)	• Durezol tid • Thera Tears SteriLid Scrub bid
One Week +5 days	• Removal of PKS • Thera Tears SteriLid Scrub bid	• Durezol bid • Thera Tears SteriLid Scrub bid
Two Weeks +5 days	• Thera Tears SteriLid Scrub bid	• Durezol qd • Thera Tears SteriLid Scrub bid
Three Weeks +5 days	• Thera Tears SteriLid Scrub bid • Autologous Serum Eye Drops (ASED) 6x daily	• Discontinue Durezol • Thera Tears SteriLid Scrub bid • Autologous Serum Eye Drops (ASED) 6x daily
6 weeks	• Refraction resulting in 20/15 visual acuity	• Refraction resulting in 20/20 visual acuity
3 months*	• Meibomian Gland Expression in-office • Thera Tears SteriLid Scrub bid • ASED qid	• Meibomian Gland Expression in-office • Thera Tears SteriLid Scrub bid • ASED qid
4 months *	• Durezol Taper (qid x 1 week, tid x 1 week, bid x 1 week, qd x 1 week) • Thera Tears SteriLid Scrub bid • ASED qid	• Thera Tears SteriLid Scrub bid • ASED qid
13 months **	• Thera Tears SteriLid Scrub bid • ASED qid	• Thera Tears SteriLid Scrub bid • ASED qid

** Following this appointment, the patient self-discontinued all therapy, reportedly due to feeling asymptomatic, which lead to a recurrence OD only at month 4.*
*** OD has been asymptomatic for 8 months, and OS for 12 months*

though temporary, with topical ocular steroids and antibiotics, which are the classic treatments for this condition.[4–13]

Differential Diagnosis

A few key differential diagnoses must be eliminated. While vernal keratoconjunctivitis can have similar corneal appearance,[13] there is no papillary response, and she has no history of allergies or asthma. Rosacea is known to be associated with meibomianitis, chalazia and hordeola.[14,15] The patient was not manifesting obvious meibomianitis. She also lacks any skin abnormalities, such as acne or telangiectasia of the face. While it was confirmed at a later date that her inferior meibomian glands were poorly functioning and mildly atrophic (see Figure 2), the severity of the meibomian gland dysfunction would not, alone, be causative for the severity of inflammation and infiltrates. There have been documented cases associating HLA phenotypes, HLA A-26 and HLA B-53 with the development of phylectenular keratitis.[16] Due to cost, serological testing has been deferred for this patient. We will consider exploring this type of medical laboratory testing in the future, as necessity dictates.

The presence of *Demodex folliculorum* can also not be ruled-out. Cylindrical dandruff at the lash base, as noted by Gao, et al.,[17] is highly suggestive that *Demodex* blepharitis is present. *Demodex* blepharitis can lead to similar corneal presentation, including corneal scarring, neovascularization, phlyctenule-like opacities, and corneal infiltrates.[16–18]

DISCUSSION

Common acute treatments for keratoconjunctivitis secondary to *Staphylococcus* hypersensitivity, include topical antibiotics and steroids.[4–13] Repeated use of topical ocular steroids does not come without side effects, including risk of increased intraocular pressure, early cataract formation, and delayed wound healing.[19–22] Topical ocular steroid tapering needs to be slow and controlled to prevent rebound inflammation.[19–22] Kwok, et al.[21] demonstrated that utilization of post-surgical topical steroids (dexamethasone) resulted in an increase in intraocular pressure, particularly in children younger than ten years old. Therefore, use of stronger topical steroids should be avoided in children. Softer steroids, such as fluorometholone**,** are less likely to induce this increase in intraocular pressure. Depending on the severity of the presentation of keratoconjunctivitis secondary to *Staphylococcus* hypersensitivity, a soft steroid may not be enough to control inflammation.

Commercially available cryopreserved amniotic membranes, such as the PKS utilized in this case, are well documented in the treatment of inflammatory and infectious corneal disease. These membranes contain both anti-inflammatory and antimicrobial properties due to the HC-HA-PTX-3 molecule inherent within.[23] Prokera amniotic membranes are placental-derived from live tissue donors following cesarean section births. The tissue is then decellularized and cryopreserved. Prokera amniotic membranes are indicated in mild to severe corneal inflammation and infection.[24–27] The wear time for this treatment is five to tendays.[25] Cheng et al[25] demonstrated that a five daywear time for the treatment of moderate dry eye disease resolved all cases of superficial punctate keratitis in 15 eyes in this study. This treatment is FDA-regulated and approved. Cryopreserved amniotic membranes offer a novel approach to treating superficial punctate keratitis without the known side-effects from topical steroid utilization.[19–22]

Once the inflammation has been reduced, the secondary goal in treatment of keratoconjunctivitis secondary to *Staphylococcus* hypersensitivity is to prevent recurrence. Tea tree oil-based lid scrubs help to diminish both *Staphylococcus* and *Demodex follicularis* species.[28–31] It has also been demonstrated that tea tree oil does not result in the generation of resistant microbial species.[28,29] Patients with *Staphylococcus* hypersensitive keratoconjunctivitis are in need of constant reduction of these species, so tea tree oil based lid scrubs offer a potential option for these patients. Topical antibiotics, even those without preservatives, can have cytotoxic effects on the cornea.[32,33] While tea tree oil can also be cytotoxic to the cornea, the therapeutic concentration in commercially available lid scrubs does not reach these cytotoxic levels.[34]

Mild residual inflammation can be reduced with the daily utilization of a chronic topical inflammatory agent. Autologous Serum Eye Drops (ASED), compounded from the patient's own blood serum, contain similar components to natural tears (vitamin A, vitamin E, fibronectin, epidermal growth factor, transforming growth factor –β, platelet-derived growth factor, fibroblast growth factor, hepatocyte growth factor, substance P, insulinlike growth factor, and nerve growth factor).[35–37] Additionally ASED contain IL-1 receptor agonists which are anti-inflammatory.[35] These drops are void of irritating and dry eye-inducing preservatives, as the blood serum is diluted with sterile saline. Concentrations of 50–100% autologous serums are recommended for moderate to severe dry eye.[36–39] ASED are safe to use daily long term. Bacterial contamination and preservation of the components in ASED is not a concern provided the ASED are cold stored properly (unopened vials for six months in the freezer or open vials for seven days in a refrigerator).[37,39] Typical dosing is between six times daily to every two hours for moderate to severe dry eye.[35,37] ASED are only contraindicated in patients with blood-borne pathogenic diseases such as human immunodeficiency virus or hepatitis B.[37] Ocular surface dryness is a possible result of chronic or recurrent inflammation. The Dry Eye Workshop

(DEWS II) Report has linked chronic inflammation as contributory to dry eye.[40] Patients such as the case subject presented here could develop dry eye symptoms as a result of these episodes of chronic recurrent inflammation, even with successful treatment of blepharo- keratoconjunctivitis. ASED offer an option for chronic dry eye therapy as well.[35,37,41]

CONCLUSION

In this patient, a topically placed, cryopreserved amniotic membrane resulted in a faster treatment time course to reduce inflammation (2 weeks) than a classic steroid taper (3.5 weeks). The eye treated with an amniotic membrane also resulted in superior visual acuity and comfort following treatment with fewer potential side-effects than the classic steroid taper. Tea tree oil based lid scrubs and autologous serum eye drops worked effectively in this patient to prevent recurrence, provided that they were used daily. Further comparison and study of the treatment modalities above could result in potentially faster, safer, and superior treatment and prevention protocols for topical inflammation in *Staphylococcus* hypersensitive keratoconjunctivitis.

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