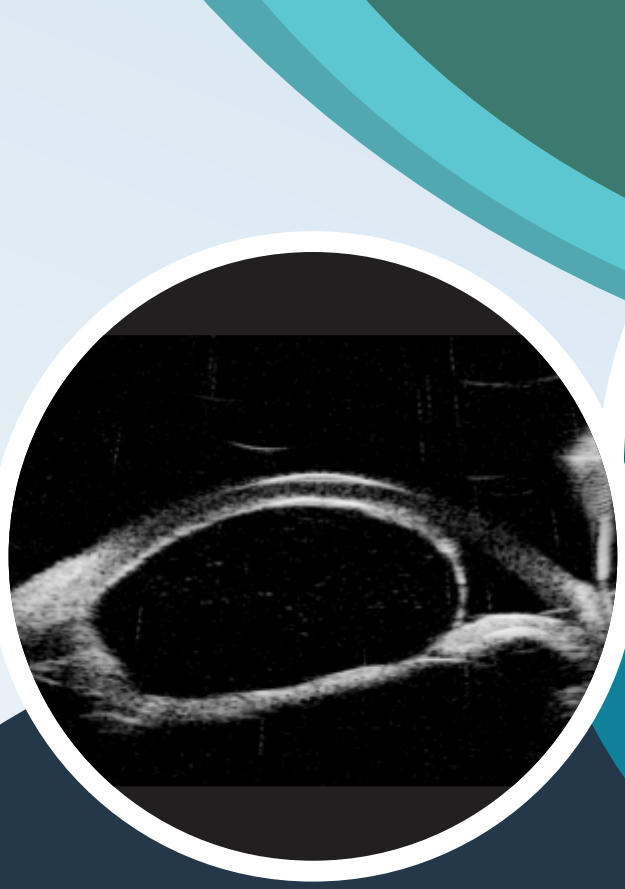
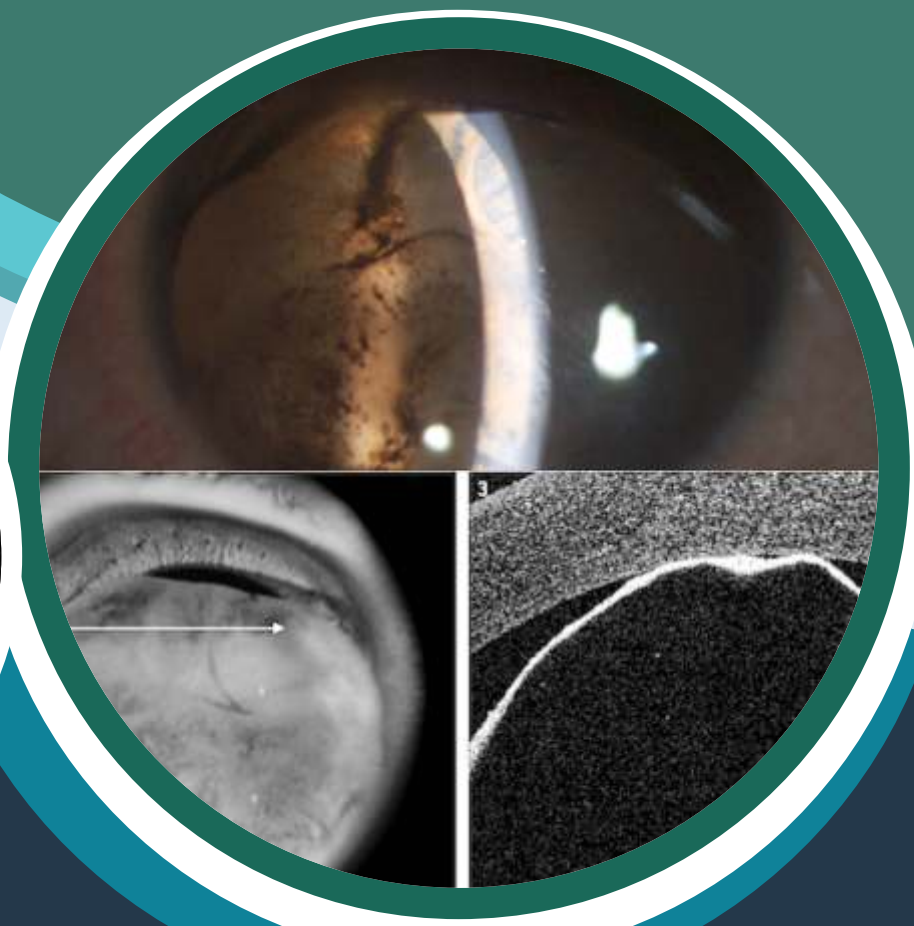
KeraSight

Official Publication of Indian Society of Cornea and Keratorefractive Surgeons

Volume 3; January - March 2018





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Volume 2; July - September 2017

Editorial

Dear Friends,

It gives me immense pleasure to present to you the July-September issue of the "KeraSight". In this issue the first article is "In-Focus" which is dealing with the antibiotic prophylaxis for cataract surgery and the spectrum of polymers for dry eye disease. ISCKRS had organized a CME in May 2017 in which a detailed discussion was done on the regimen of antibiotic prophylaxis in cataract surgery and how long and how frequent should an antibiotic drop be put before and after cataract surgery. The role of intracameral antibiotics at the end of the surgery, its pros and cons were also discussed. Similarly the role of topical steroid, its frequency and duration after cataract surgery and how and till what time it should be used and how should it be tapered was discussed. The role, advantages and disadvantages of combination drops of antibiotic and steroid were also highlighted. Apart from these an interesting discussion was done on the diagnostic dilemmas of dry eye and allergic eye disease and the available polymers for use in dry eye disease. We had a unique format of 5 minutes of presentation and 15 minutes of discussion after each presentation which was very much appreciated by all the delegates. In this format we had a group of panelists apart from the chairpersons and hence a very useful and meaningful discussion was possible. The first article is a compilation of the opinions given by the speakers and the panelists.

The second annual conference of the ISCKRS- "ISCKRS MEET 2017"- will be held on the 5th & 6th of August 2017 at India Habitat Centre, Lodhi Road, New Delhi. The theme of the Annual Conference this year is 'Cornea & Keratorefractive: Newer Paradigms'. It gives me immense pleasure to invite you to the Annual Conference of ISCKRS which will bring together practitioners, researchers and educators from India and abroad who are engaged in state of art academic work in cornea and Refractive Surgery. It will be a two-day event that will involve Live surgery demonstration, Orations & key-note lectures, Sessions on Corneal problems and their solutions, Refractive surgical issues, Video assisted sessions, sessions for Young Ophthalmologists, and free paper sessions. Hands on wet labs and skill development sessions will be provided by experts in contact lens and phacoemulsification. The ISCKRS SightLife wet lab on DSAEK and DALK will be a special attraction for the delegates. A special "ISCKRS Ophtha Quiz" will be organized which will allow everyone present in the hall to participate and win. The interest that our fellow ophthalmologists are showing in this event is very encouraging and I am sure the conference is going to be very successful.

Hoping to see you in the conference.

Rajesh Sinha

Rajesh Sinha
MD, DNB, FRCS
Gen. Secretary ISCKRS

INDIAN SOCIETY OF CORNEA
AND KERATOREFRACTIVE SURGEONS

KeraSight

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In Focus:

Vital Dyes For Keratorefractive Surgeons

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Introduction

Dyes are chemical compounds that bind to various substances in nature to induce colour. When dyes colour living tissues or cells, they are called vital dyes. Vital staining refers to the coloration of living cells or tissues. When vital staining is done in a living organism, it may be called intravital staining, whereas supravital staining is defined as the application of dyes to living cells or tissues freshly removed from the body. Vital dyes emerged as important and effective surgical adjuvants to enhance visualization of ocular tissues.

Commonly used vital dyes used by keratorefractive surgeons are as follows.

Trypan Blue (TB)

TB is an anionic hydrophilic azo dye with the formula $C_{34}H_{24}N_6Na_4O_{14}S_4$ and a molecular weight of 960 daltons. Azo dyes are large class of synthetic organic dyes that contain nitrogen in the azo form (-N 5 N-) in their molecular structures connecting aromatic ring compounds. Live cells/tissues with intact cell membranes usually are not coloured because their selective control of cellular membrane transport does not allow binding of TB. It has been widely used in both vitrectomy and cataract surgery and is commercially available in an 0.15% concentration for vitreoretinal surgery under the brand name Membrane Blue (DORC International, Zuidland, Netherlands) and as Vision Blue in a 0.06% concentration for cataract surgery (DORC International). TB as Membrane Blue and Vision Blue comes in a solution containing small amount of sodium salts, 8.2mg of NaCl, and water. The osmolarity of Vision Blue ranges from 257 to 314 mOsm/Kg and the pH from 7.3 to 7.6.

USES

TB has been used in many eye banks to evaluate corneal endothelial cell viability. The blue dye stains the nuclei of damaged and dead endothelial cells in donor corneas, as well as areas of DM denuded of endothelial cells. In order to assess endothelial damage and endothelial cell density by light microscopy, Sperling in 1986 introduced the use of TB diluted in 0.45% and 0.9% sodium chloride, or with 1.8% sucrose to induce dilation of intercellular spaces for visualization of endothelial cell borders. However, the reliability of TB-guided endothelial cell counts has been questioned. Although there was a significant correlation between the techniques, TUNEL-assay seemed to be more reliable in detecting earlier events in endothelial death process. At the frequently used concentrations of 0.001--0.1%, TB is not toxic to the corneal endothelial cells. Further investigation may clarify the benefits of TB-guided endothelial cell assessment for clinical practice.

TB is used in both penetrating and deep lamellar keratoplasties. For penetrating keratoplasty, 0.02% TB solution may be injected into the anterior chamber via a paracentesis to stain the DM of the donor as well as the recipient corneal tissue. Dye exposure may promote close alignment of edges of host and donor DM, thus improving graft stability and minimizing surgically induced astigmatism. TB may show both cut edges well, identify the corneal depth during suturing; and enable visibility of the viscosurgical device removal. Thereby the blue dye may improve tissue apposition.

TB has been also used in order to facilitate stromal dissection and to avoid DM perforation in lamellar keratoplasty. After a two-thirds trephination of the cornea, 0.02% TB solution is injected intrastromally in four quadrants through a 30-gauge cannula for superficial dissection. Next, more TB is injected for deep dissection to stain the stromal fibers. This novel approach resulted in no residual stroma at the end of the intervention. TB disappeared totally in the early postoperative period.

Removal of DM during endothelial keratoplasty i.e. DMEK, DSAEK/ DSEK in pseudophakic eyes is greatly felicitated by staining the host DM with TB. (Fig 1&2)

It is also used to remove retained DM after penetrating keratoplasty as in congenital hereditary endothelial dystrophy because of longstanding stromal edema and loosening of the attachment. In this case, the simple, quick application of TB facilitates the removal of retained membranes during penetrating keratoplasty. In summary, the TB stain may be useful intra-operatively to visualize and remove the posterior stromal layers in modern keratoplasty.

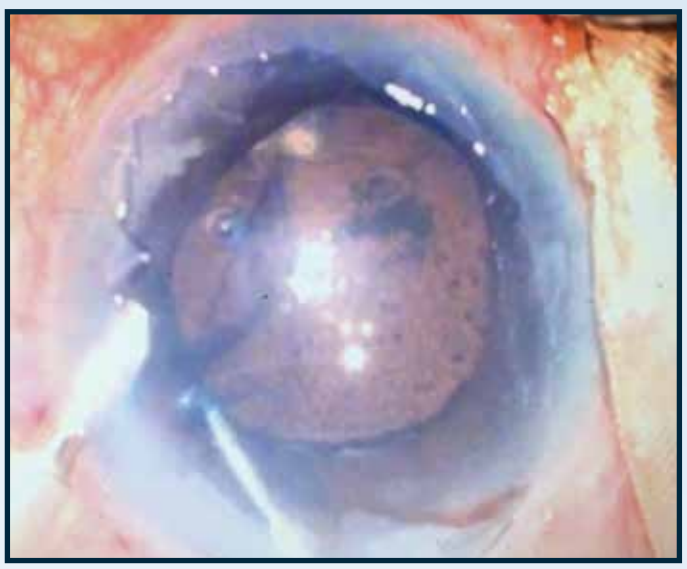

Figure 1. *Removal of trypan blue stained DM during descemetorhexis during DSAEK.*

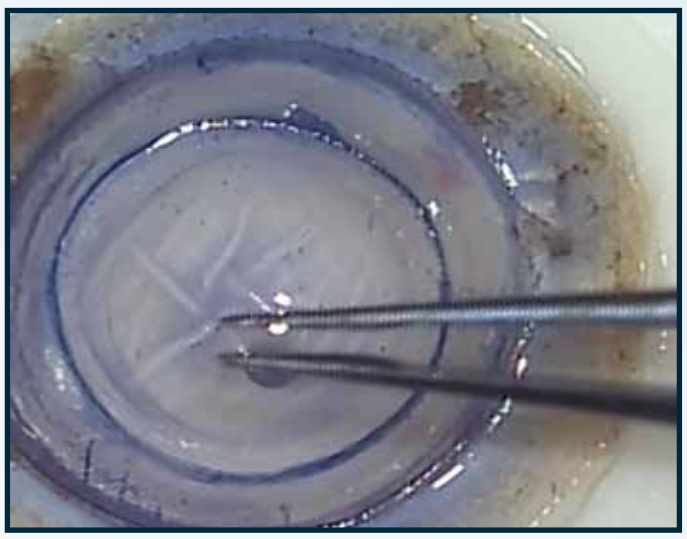

Figure 2. *DMEK lenticule preparation using trypan blue.*

Due to the frequent difficulty in finding clear corneal incisions during intraocular surgery, the use of TB-coated blades has been proposed. A 3.0-mm phaco incision blade tip was coated with TB in order to improve visualization. Fluid outflow and the mechanical effect of phaco tip movement during surgery also facilitated visualization of the incision, and no complications were reported with this surgical method. The vital dye had disappeared from the cornea after few days.

TB staining of the capsule edge in CCC was initially introduced in 1999. Since then the blue azo dye has been the most frequently used agent for staining the anterior capsule, as intra- operative injection of TB provides various advantages in cataract surgery First, surgical TB injection may promote higher rates of success of capsulorhexis in phacoemulsification for cases with inadequate red reflexes. The rate of conversion to an extracapsular cataract extraction in white cataracts as the result of an incomplete CCC has been as low as 3.85% when TB is used, compared to 28.3% when no dye has been injected. Second, the blue vital stain allows staining of anterior and posterior capsules in children younger than 5, thereby enhancing effectiveness for completing the CCC. Thirdly, the importance of TB in the learning process of trainee surgeons and residents to perform CCC has been recently highlighted. Even in the presence of a good red reflex, young surgeons had a higher success rate of CCC when the dye was used. Finally, 0.3 ml of 0.06% TB has been applied to find the edge of a lost capsulorhexis, allowing the CCC completion.

The positive effect of TB staining of the lens capsule is that it may modify the biomechanical structure. Wollensak and co- authors recently performed a study on porcine eyes that evaluated 0.1% TB stain for 30 seconds, 1 minute, and 30 minutes with or without white light. TB staining for 1 and 30 minutes led to an increase in elastic stiffness and a significant decrease in ultimate extensibility in the anterior capsule. This effect was probably due to the photosensitizing action of TB to physical cross-linking of collagen through yielding of free oxygen radicals, which causes a change in elastic behavior.

Recently, Singh et al analyzed histological characteristics of TB-stained human capsules. They found out that TB mostly stained the basement membrane adjacent to the epithelial layer of the lens capsule with minimal laminar staining in the

superficial basement membrane. Such findings may also explain why TB may provide differentiation between non-stained cortex and the lens capsule.

Toxicity

Low doses of TB do not produce inflammation and corneal toxicity when injected into the anterior chamber. Clinically, TB to facilitate CCC is used in concentrations from 0.0125% to 0.4%. One uncontrolled case series where 0.1% TB was instilled into the anterior chamber found no effect on endothelial cell count or pachymetry during 8 years of follow-up. Another randomized clinical trial found 7.5% endothelial cell loss when 0.06% TB was placed under air bubble, compared to the 10.2% loss in the group with only air. In a rabbit model TB showed no corneal endothelial cytotoxicity after 1minute exposure even at a concentration of 0.4%. TB placed under an air bubble or under viscolelastic produced no toxicity in the follow-up period. TB staining of the anterior capsule may not result in corneal toxicity (Table 1B).

Density and viability of lens epithelial cells (LEC) predicts posterior capsule opacification after cataract surgery. Nanavaty et al used 0.0125% TB to perform CCC in patients with white cataracts, and a sample of the anterior capsule was excised and compared with non-stained capsules. A significant decrease in LEC density was detected in eyes treated with TB compared with untreated ones, suggesting a toxic effect to the LEC. On the other hand, Melendez et al found neither toxicity nor photosensitization in cultured LEC with concentrations of TB ranging from 0.025 to 4mg/ml. Further studies are needed.

A relative contraindication for TB is the use of hydrophilic expandable acrylic intraocular lenses (IOLs), which have the highest water content (73.5%) of currently manufactured implants. This IOL is implanted dry, and its expansion depends on hydration by fluids in the capsular bag. Because the TB that remains in the aqueous humour may be absorbed into the lens, patients may experience more glare than with other IOLs; however, no significant difference was found in contrast sensitivity.

Bascal et al report a case where an area of zonulysis allowed 0.06% TB to enter the vitreous cavity. They observed a moderate bluish hue of the vitreous that persisted until the fourth day. Multifocal ERG showed reduced responses which normalized after one month from the surgery, indicating a transitory retinal toxicity.

Fluorescein

Fluorescein is a xanthene fluorophore with chemical structure of C20H12O5 and molecular weight of 332 daltons. The vital dye in water has a very high fluorescence with an absorption maximum and excitation at 494 nm and emission maximum of 521 nm. Fluorescein may be conjugated with over 50 different salts or derivatives, including fluorescein sodium (FS) and fluorescein diacetate (FD). Fluorescein derivatives have been commonly used in microscopy, in dye laser as the gain medium, in serology to detect latent blood stains, and in dye tracing. Fluorescein is used extensively as a diagnostic tool, mainly as FS. (Fig 3)

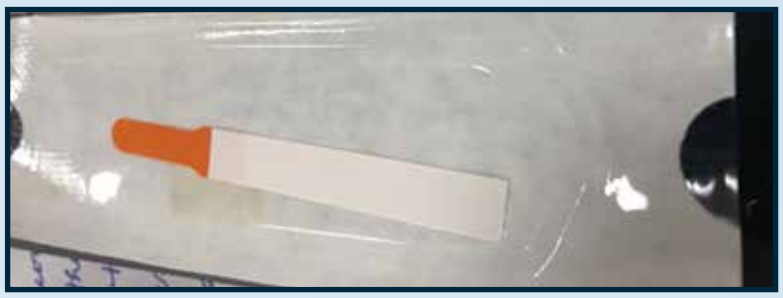

Figure 3. *Fluorescein strips*

Corneal Staining

Fluorescein doesn't penetrate intact corneal epithelium. But if there is a break in the epithelium, the fluorescein penetrates in to exposed Bowman's membrane and stroma and makes contact with alkaline interstitial fluid and turns in to bright green owing to its pH indicator properties. It is best examined under cobalt blue filter of the slit lamp and is very helpful in identification size of the lesion as well as occult lesions. Common staining patterns seen in the cornea are as follows

- Interpalpebral: dry eye
- Horizontal band across inferior 1/3 of cornea: lagophthalmos /exposure
- Superior punctate: SLK, floppy eyelid syndrome
- Central punctate: focal epithelial keratitis (Thygeson's SPK, epidemic keratoconjunctivitis, molluscum)
- Inferior punctate: blepharitis
- Perilimbal (360°): soft contact lens wear

Seidel's test: A drop of fluorescein is instilled into the lower fornix and patient is asked to blink to

spread the dye evenly. On examination with slit lamp using cobalt-blue filter, fluorescein will be diluted by aqueous at the site of leakage

Applanation tonometry: After anaesthetising the cornea with a drop of 2 per cent xylocaine and staining the tear film with fluorescein patient is made to sit in front of slit-lamp. The cornea and biprisms are illuminated with cobalt blue light from the slit-lamp. Biprism is then advanced until it just touches the apex of cornea. At this point two fluorescent semicircles are viewed through the prism. Then, the applanation force against cornea is adjusted until the inner edges of the two semicircles just touch. This is the end point. The intraocular pressure is determined by multiplying the dial reading with ten.

Contact lens fitting assessment: Fluorescein allows us to evaluate the physical fit of a contact lens, or the bearing relationship that exists between a transparent contact lens and the eye. A yellow-green glow indicates the presence of fluorescein and therefore clearance between the lens and cornea. A deep blue or black area indicates the absence of fluorescein and therefore the lens is touching the cornea. Between these two extremes, the varying thickness of the tear layer is observed by varying shades of green. Bubbles indicate an air space and appear black.

Tear film BUT: It is the interval between a complete blink and appearance of first randomly distributed dry spot on the cornea. It is noted after instilling a drop of fluorescein and examining in a cobalt-blue light of a slit-lamp. BUT is an indicator of adequacy of mucin component of tears. Its normal values range from 15 to 35 seconds. Values less than 10 seconds imply an unstable tear film.

FDDT: Instill fluorescein in inferior fornix of both eyes, wait 5 minutes, then evaluate for asymmetric clearance of dye from tear meniscus. A prolonged retention of dye in conjunctival sac indicates inadequate drainage which may be due to atonia of sac or mechanical obstruction.

Jones Dye Test:

- **Jones I test:** perform dye disappearance test, then attempt to recover fluorescein in inferior nasal meatus with cotton-tipped applicator after 5 minutes; abnormal result (no fluorescein) occurs in 33% of normal individuals. If the dye is retained in cotton-tipped applicator, the cause of tearing is probably hypersecretion i.e. epiphora.
- **Jones II test:** perform Jones I test, then irrigate saline into nasolacrimal system; dye recovery from nose indicates functional occlusion of nasolacrimal duct; clear saline recovery indicates canalicular occlusion or nonfunctioning lacrimal pump. (Fig 4)

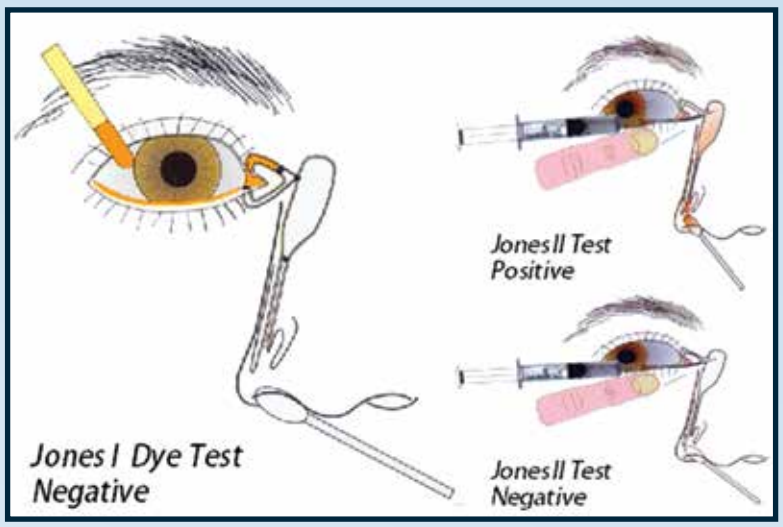

Figure 4. *Jones 1 and 2 tests.*

Rose Bengal

Rose Bengal (RB) is the 4,5,6,7-tetrachloro-2′,4′,5′,7′-tetraiodo derivative of fluorescein and has been widely used to detect damage to the ocular surface epithelium (mainly on the conjunctiva, due to poor contrast on the cornea) (Fig. 5). Traditionally RB has been thought to stain dead or desquamated, but not healthy epithelial cells; this concept was introduced in the 1930s by Sjögren and later reiterated by Passmore and King, but no direct evidence was ever provided. In vitro studies on human corneal-limbal epithelial cells have since demonstrated the important role of membrane-associated mucins (in addition to secreted mucins) in blocking RB staining. It is likely that in patients with dry eye RB staining occurs in areas lacking these mucins. Positive staining of the cornea with Rose Bengal indicates a qualitatively and/or quantitatively inadequate tear film. Hence this diagnostic staining is used particularly if there is suspicion of keratoconjunctivitis sicca or there is too rapid disruption of the tear film without epithelial defects (fluorescein-negative). The number of purplish-red flecks, which can only be counted carefully with the aid of a slit lamp microscope, is a measure of the loss of cells and consequently of the severity of the disturbance of the tear film.

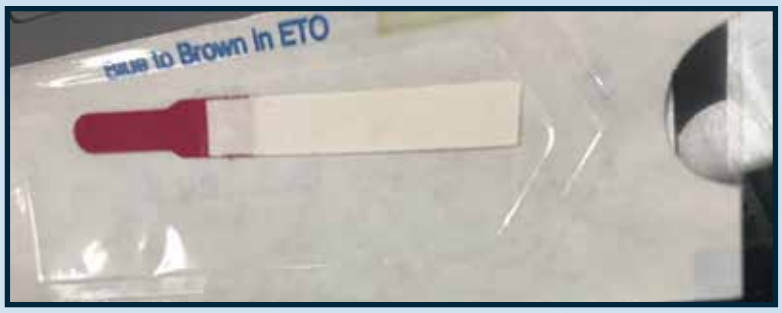

Figure 5. *Rose Bengal strips*

The stain is irritating to the eye, so a local anesthetic should be applied in advance, and when defects are present the staining is very persistent. Feenstra and Tseng also highlighted the intrinsic toxicity of RB, which induces an instant loss of vitality (as shown by immediate cellular morphologic changes, subsequent loss of cellular motility, cell detachment and death). This intrinsic toxic effect is augmented by light exposure.

Rose Bengal has also been shown to stain the ocular surface in a variety of diseases that may not necessarily be associated with dry eye. In conjunctivochalasis RB staining can be observed along the redundant conjunctival folds, the area above these folds, the adjacent lid margin and the conjunctival non-exposure zone. In lipid tear deficiency caused by noninflamed meibomian gland dysfunction RB staining preferentially occurs in the non-exposure zone, while in superior limbic keratoconjunctivitis it can be detected in the superior bulbar conjunctiva. In LASIK-induced neurotrophic epitheliopathy RB staining typically affects the cornea. Finally, RB staining can be detected in atopic keratoconjunctivitis, herpes simplex keratitis and ocular surface neoplasia.

Lissamine green staining

Lissamine green (LG) is a synthetic organic acid dye with two aminophenyl groups. that stains the ocular surface similarly to RB without causing stinging. Staining is dose dependent and a minimal dosage of 10–20 μl is recommended, since inadequate volume results in a weak staining pattern that can be overlooked or underestimated. When LG is used, attention should be paid to the timing of observation, because an overly hasty evaluation is likely to hamper a full understanding of the staining pattern (evaluating the staining too quickly does not allow full development of the staining pattern and delay in evaluating the stain results in fading of the pattern). Ideally the grading should be performed between one and four minutes after staining. For a better reading it is also essential not to use an intense illumination beam, which may reduce the contrast and lead to an underestimation of grading.

In the clinical setting, the staining profile of LG is nearly identical to that of RB and they are considered to be interchangeable. However, experimental studies have detected important differences between LG and RB: the former, for example, does not stain healthy corneal epithelial cells, nor does it affect their viability, whereas both characteristics are typical of RB.

Gentian Violet (GV)

GV is used to mark the peripheral stromal surface containing DM and endothelium in cases of DSAEK/DSEK and DMEK. By identifying the side of the marked cornea, surgeons may insert the folded donor lenticule into the host anterior chamber in the proper position. This ensures that air is injected posterior to the graft, thus preventing the donor lenticule from unfolding upside down.

It is also used to mark the host cornea for proper centration before trephination and suture placement marks during DALK and penetrating keratoplasty.

Nocardia Scleritis – Clinical Presentation and Management

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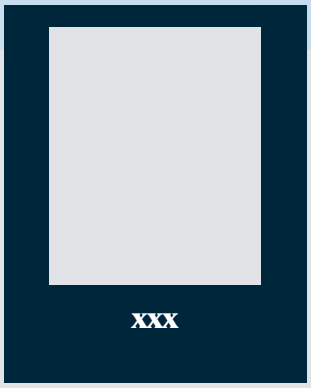

Keywords: Nocardia, necrotizing scleritis, infectious sceritis, scleritis

Abstract

Aim: To report an unusual case of Nocardia scleritis and present an updated review of literature.

Methods & Results: A 62 years old male diabetic patient was diagnosed with left eye necrotizing scleritis unresponsive to steroids. Microbiology revealed Nocardial infection. Prolonged and intensive medical management with topical fortified 2.5% amikacin and 0.5% moxifloxacin led to the resolution of scleritis.

Conclusion: A high index of suspicion of infection by the treating ophthalmologist and prolonged intensive medical management can significantly optimize the outcome in rare cases of Nocardia scleritis.

Introduction

Necrotizing scleritis is a chronic destructive inflammatory disease of the sclera, which if left untreated can progress to perforation and possibly even blindness. In almost 40-90% of the patients of necrotizing scleritis there is an underlying autoimmune disease, Rheumatoid arthritis being the commonest of all [1-3].

Infection scleritis constitutes just 5-10% of scleritis cases [1, 2, 4]. Nocardia is a rare cause of infectious scleritis. Nocardia keratitis and sclerokeratitis is commonly reported, but its presentation as an isolated scleritis has rarely been reported. We report this rare presentation of Nocardia scleritis and describe the various clinical presentations and management of ocular nocardial infections.

Materials and Methods

A 62 years old male patient, farmer by profession, presented with chief complaints of gradually progressive, painful diminution of vision in the left eye for last two months associated with redness, watering and headache. Patient was diabetic and on irregular oral hypoglycemic medications. He was diagnosed as a case of necrotizing scleritis by a private practitioner and medically treated with oral and topical steroids that resulted in deterioration of symptoms.

On examination, best-corrected visual acuity (BCVA) was 20/40 in both eyes. Ocular examination in left eye revealed two punched out inferior scleral

lesions, measuring 4.5x2.8mm and 2.5x2mm respectively covered with whitish necrotic debris with underlying scleral. Two other similar lesions measuring 1x1.5mm were seen in the nasal and temporal sclera. Cornea was clear with a quiet anterior chamber. Posterior segment was normal in both the eyes (Figure 1).

Figure 1a. *Punched out sclera lesions with necrotic debris*

Fasting blood sugar level was 151mg/dl. Serum biomarkers (for autoimmune diseases) were negative. Chest X-ray revealed a normal study. Scleral exudate was sent for microscopic examination (Gram stain, KOH stain, modified Zeihl-Neelson stain using 1% H2SO4) and culture (blood agar and Sabouraud dextrose agar). Staining revealed thin, beaded, branching filaments that were gram positive and weakly acid fast. Culture on blood agar showed tiny, dry chalky white colonies suggestive of Nocardia species (Figure 2).

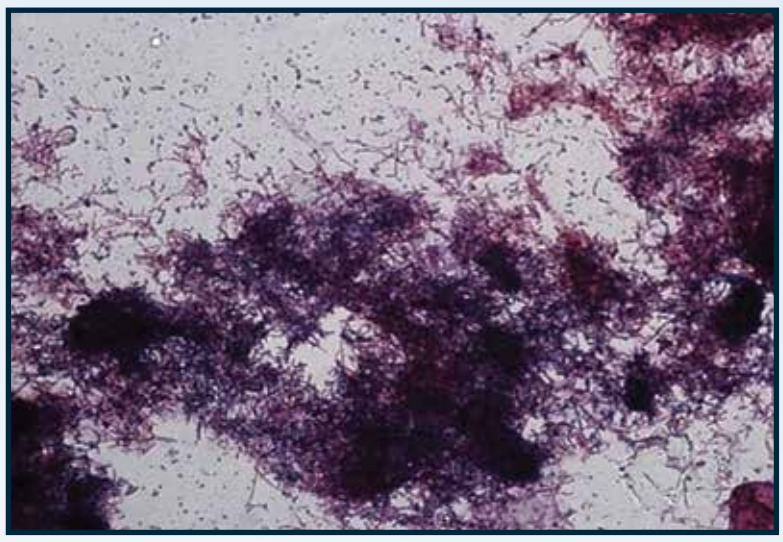

Figure 2b. *Thin beaded branching gram positive filaments on Gram's stain*

Based on antibiotic sensitivity reports patient was started on topical amikacin 2.5% and topical moxifloxacin 0.5% one hourly, topical cycloplegics 6 hourly and oral ciprofloxacin 500mg twice daily that was tapered over next three months.

Results

After an initial response to treatment, patient had a recurrence of pain, diminution of vision (visual acuity of 20/200) associated with proptosis of 4mm (by Hertel's exophthalmometer) in the treating eye. CT scan head and orbit showed posterior scleritis, suggestive of recurrence of infection (Figure 3A). Fundus was normal on examination. The patient was again started on frequent topical fortified antibiotics- 2.5% amikacin and 0.5% moxifloxacin- and systemic antibiotics for a prolonged period of six months with a more gradual tapering to avoid any further recurrence. At six months, patient achieved BCVA of 20/40 with the healing of the lesion.

Discussion

Infection constitutes a rare cause of necrotizing scleritis. Often the condition is associated with an underlying autoimmune disease that primarily requires treatment with steroids. In case an underlying infection is missed, as was seen in our case, this may worsen the disease. Hence, it is imperative to rule out infection in doubtful cases, and practice caution before starting steroids.

Pseudomonas aeruginosa is the most commonly reported organism worldwide in infectious scleritis [5-7] whereas in Indian scenario, Aspergillus Flavus and Staphyloccus have been reported to be the leading cause. Nocardia though a rare cause of scleritis worldwide appears to be commoner in Indian scenario [8, 9]. This is possibly because of higher prevalence of Nocardia in Indian soil and greater risk of exposure to dust in an agricultural country like India [10]. The various predisposing factors in our patient were uncontrolled diabetes, agricultural profession that might have exposed the patient to some trivial trauma that he ignored and inappropriate steroid usage.

Nocardia is an aerobic, gram positive, weakly acid-fast, branching filamentous bacteria that belongs to the order Actinomycetales. A saprophyte in soil, Nocardia is not a part of normal ocular flora. Though systemic nocardiosis has been associated with immunocompromised status, ocular infections like keratitis, scleritis, orbital cellulitis, and endophthalmitis [11] have been commonly reported in immunocompetent individuals. Keratitis is the commonest ocular nocardial infections typically presenting as a patchy anterior stromal infiltrates in a 'wreath' pattern or raised pinhead lesions. Scleritis

usually occurs as an extension of keratitis with areas of well circumscribed punched out lesions [12]. Clinical presentation of our patient as isolated nocardia scleritis is rare.

The diagnosis is straightforward by staining and culture. But, availability of a microbiology set-up is a limiting factor in developing countries leading to a delay in diagnosis and subsequent complications like perforation. The management is mostly medical based on the culture and sensitivity reports. The sensitivity reported is 100% with topical fortified amikacin (2.5%) and topical trimethopim (16mg/ml)-sulfamethoxazole (80mg/ml) and intermediate sensitivity for fluoroquinolones, cephalosporins and tobramycin. Amikacin is considered the drug of choice [13]. In our patient, the organism was sensitive to amikacin and moxifloxacin and responded to the same. Surgical intervention in the form of debridement may be needed along with the medical management [8]. A prolonged intensive treatment for atleast 4 to 6 months is essential for complete resolution and prevention of recurrences.

In conclusion, a high index of suspicion of infection by the treating ophthalmologist, avoiding inappropriate corticosteroid usage and a timely referral to a tertiary eye care center can significantly optimize the outcome even in rare cases of Nocardia scleritis.

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Post Surface Ablation Haze

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Introduction

Refractive Surgeries like Photorefractive Keratectomy (PRK), Laser-assisted Subepithelial Keratomileusis (LASEK) and Epi-Laser in situ Keratomileusis (Epi LASIK) have one thing in common that they all use surface ablation in some form and this may be the reason that they produce post-operative corneal haze. In all three procedures, laser ablation with excimer laser is done after removal of corneal epithelium but with different techniques. In PRK, epithelium is removed mechanically with micro instruments, in LASEK it is ethanol-assisted and in Epi-LASIK epikeratome is used to separate epithelium. In LASEK and Epi-LASIK epithelium is reposited back after ablating corneal stroma but in PRK epithelial healing occurs by primary intention as there is no epithelial flap. This may be the reason that post-operative haze is mostly seen with PRK (Figure 1). Subepithelial haze is seen almost in all post PRK patients after 1month, reaching the greatest intensity at 3-6 months, which gradually decreases from there.[1,2,3]

Commonest risk factors accounting for increased post-operative corneal haze are:

- Myopic correction > -6.0D, Hyperopic correction > +3.5D, Astigmatism > 1.5D,
- Ablation depth > 80μm,
- Smaller diameter ablation profile,
- Retreatment / enhancement,
- Previous corneal surgery

Figure 1a. *Punched out sclera lesions with necrotic debris*

Other factors that may influence occurrence of sub-epithelial haze following surface ablation includes pregnancy, oral contraceptive use, ultraviolet (UV) radiation exposure, ocular surface disorders and those with brown irides.[4,5]

Historical Perspective and Epidemiology

The advent of Excimer laser brought the origin of corneal refractive procedures. PRK was developed by Trokel and colleagues in 1983 using excimer laser that emits ultraviolet light of 193 nm. In 1996, it was approved by Food and Drug Administration (FDA) as refractive surgery technique.[6,7] Since then thousands of people have undergone PRK world over. The PRK had limitations of post-operative discomforts in the form of pain and corneal haze. PRK was followed by LASIK which had excellent results without post-operative discomforts and with immediate visual recovery with 'wow' factor. LASIK has its own share of complications in the form of post-operative corneal ectasia and flap complications which led to resurgence of flap less refractive surgeries i.e.

surface ablation. The prevalence of post-operative corneal haze has been studied in different studies. It is variable and depends on pre-operative risk factors and the surgical approach. In a study by Netto MV et al the incidence of clinically significant corneal haze was 0.3% to 3%.[8] In another study at Singapore by Bryan chin et al, it was reported to be around 2.3 % and it was more when refractive surgery was done for higher corrections.[9]

Pathophysiology

Post surface ablation haze has been explained by exaggerated corneal healing response in the form of abnormal tissue reaction. The corneal healing process begins following epithelial injury with the release of inflammatory mediators. Activated and upregulated growth factors, cytokines and chemokines produced by epithelial cells and found in the tear film contribute to wound healing and injury response. In the absence of protective epithelial barrier, epithelial-stromal tissue interaction initiates a cascade of events leading to keratocyte apoptosis and necrosis. This is followed by proliferation of remaining keratocytes and they synthesize new extracellular matrix which causes corneal haze. These keratocytes migrate to repopulate stroma, in turn activating fibroblasts and causing myofibroblast formation. These myofibroblast are formed in response to Transforming growth factor beta (TGF ß) signalizing pathway, which is the tissue response pathway after any penetrating injury. Myofibroblasts have greater reflectivity and tend to be less transparent because of altered crystalline production. Their extracellular matrix exhibits less organization and increased density which causes increased light scattering and haze formation.[10]

In refractive surgeries with surface ablation, there is loss of epithelial basement membrane, more so in PRK, which is responsible for corneal haze post operatively. The epithelial basement membrane is believed to act as a barrier between the stromal keratocytes and the inflammatory molecules released from the tear film and epithelium. Loss of this causes increased direct exposure of stromal tissue to the inflammatory mediators released from epithelium and tear film.

Another issue with PRK is irregular surface, which has also been related to high incidence of haze. Increased dioptric corrections results in more surface irregularity, which results in increased corneal haze. Interestingly, surface irregularity has been positively correlated with increased myofibroblasts in anterior stromal tissue.[11] The mismatch of plasminogen activator- plasmin also accounts for the corneal haze due to accumulation of extracellular matrix.

Clinical Symptomatology

Post-operative corneal haze generally causes fluctuation of vision, irregular astigmatism, reduction in contrast sensitivity, difficult night vision, myopic regression and reduction in best-corrected visual acuity (BCVA).

Types of Haze

Based on symptomatology, 2 types of corneal haze have been observed.[8] The more common is transitory haze, which is found between 1 and 3 months post operatively and rarely with clinical symptoms. This resolves mostly within 1 year. Another type is late onset corneal haze which occurs between 2 and 5 months after surgery and can cause significant compromised vision, myopic regression and can last up to 3 years.[1,12] This is generally associated with high myopic ablations and rarely can cause permanent changes requiring surgical treatment.

Corneal haze can be quantified objectively as well as subjectively. Objective grading can be done through various instruments noting the degree as well as depth of the corneal haze. For this confocal microscopy and scheimpflug densiometry is used.[13] Anterior segment optical coherence tomography (ASOCT) can also be used to measure the haze when surgical management is planned.

Subjective grading can be done based on slit lamp findings. Various investigators have devised their grading based on clinical details noted. Fantes and colleagues described 5 stages of haze based on visualization of the iris/lens details with completely clear cornea graded as grade 0 and opacification of stroma with entirely obscured view of anterior chamber graded as grade 4.[14] Similar grading system has been given by Hanna et al based-on visualization of iris details by slit lamp.[15]

Management

Management of post surface ablation haze includes prevention or medical treatment as well as surgical modalities of treatment. Prevention is aimed at decreasing corneal stromal inflammation as well

as fibrosis. As there are more chances of corneal haze after higher ablations, the role of prevention comes into play in these cases. Various modalities of preventive techniques have been described in the literature. Out of which, most practiced one is use of intra-operative Mitomycin C (MMC) and post-operative topical steroids.

MMC is an antineoplastic antibiotic agent of the family of anti-tumour quinolones, derived from Streptomyces caespitotus. It is a potent DNA crosslinker and acts in a dose dependent manner. MMC inhibit cell mitosis, including epithelial and stromal cells. MMC is used widely in other ophthalmic conditions like conjunctival neoplasms, pterygium and glaucoma surgeries to prevent fibrosis, scarring and neoplasm recurrence. It is generally used prophylactically in high risk cases which are known to have post ablation corneal haze i.e. high myopia and deeper ablations more than 80μm. The concentration of MMC, used is 0.02% and is applied for a duration of 30 seconds to 2 minutes after ablation.[3,6,16]

Although the application of MMC is beneficial for corneal recovery, it is necessary to control the doses and the time of exposure. Thornton et al, in one of studies showed that concentration is a more important factor than the duration of MMC exposure in corneal haze prevention. He advocates that for high myopia corrections (>-6.00D) standard concentration of topical MMC (0.02%) may be used, whereas for moderate myopia (-3.00 to -5.90D) low dose of MMC (0.002%) may be considered.[17]

Rajan et al analyzed the effects of MMC after correction of -9.00D by PRK in 3 groups: without MMC application, with MMC (0.2mg/ml) application for 1min and with MMC (0.2mg/ml) application for 2 min. The 2 min MMC group had thinner epithelium than the 1min and without MMC application groups.[18]

Fazel et al found that two-step administration of 0.02% MMC (45 seconds, followed by 15 seconds) further decreased corneal haze formation in high myopia, compared to a single dose of 45 seconds.[19] Shojaei et al found that short-time MMC exposure prevents low-grade haze in low ablation depths. The MMC doses oscillate between 0.002% and 0.06%. The intraoperative application of 0.02% MMC solution is the most recommended, as it produces less corneal haze, and provides better uncorrected visual acuity and best spectacle-corrected visual acuity.[20]

In one of the studies by Majumdar et al, a prospective randomized trial, 60 eyes were included for ablation with refractive error ranging from -6.00D to -10.00D. MMC treated eyes had minimal haze whereas control eyes had significant haze in almost 63% patients.[21] Mitomycin C was used in dose of 0.02% for 2 minutes. Few studies have compared the effectivity of MMC in dosage of 0.02% versus 0.01% in moderate myopia. Ramjoo et al found similar refractive and haze outcomes with 0.01% and 0.02% MMC for moderate myopia, recommending the use of 0.01%.[22]

Though MMC is effective in preventing post ablation haze, its use is not without complications. Various complications have been reported in literature i.e. ocular surface toxicity including stem cell defect, sterile melting and delayed epithelialization.[23] Although unusual, scleral ulceration and non-healing conjunctival defects have also been reported with high MMC doses (0.04%). As MMC is applied in the stromal bed, it seems that it might penetrate the anterior chamber, because cytotoxic effects on the ciliary body epithelium have been reported.[24] There has been reports of toxic effect of MMC in the overall morphology of the endothelium. Morales et al found that intraoperative 0.02% MMC applied for 30 seconds after PRK induced corneal endothelial cell loss.[25] Diakonis et al applied MMC for 15 seconds and the density of endothelial cells was not affected.[24]

Because of these potential complications, use of MMC should be restricted to the central cornea and surface of the eye and fornices should be thoroughly irrigated following application. Apart from use of MMC, other agents have been used to prevent post ablation haze. Use of topical steroid within first 72 hours is supposed to prevent haze formation.[1] For this purpose, topical prednisolone or soft steroid i.e. lotoprednol or fluorometholone has been used, however it should not be used for longer duration to curtail the risk of steroid responders. Non-steroidal anti-inflammatory drugs (NSAID) have also been used in place of steroid in few studies but results are not as good as steroids.[6]

Use of high dose vitamin A (25000IU) and vitamin E has been suggested in initial 3 months post ablation to decrease incidence of haze. They promote reepithelialization by reduction of free oxygen radicals. Prophylactic use of Vitamin C (ascorbic acid) in dosage of 1000 mg daily has been studied to prevent haze as they are supposed to reduce

keratocyte activation, impede UV damage and prevent formation of free oxygen radical.[1]

New modalities of treatment are being investigated for treatment of corneal haze. Role of plasma rich in growth factors (PRGF-Endoret) has been investigated and has shown good results.[26] Another promising result has been seen with topical interferon alpha-2b which has anti-scarring effects.[27] There is definitive role of TGF-beta in causing post-operative haze and to counter act its effect, role of anti-TGF drugs i.e. Trichostatin A is being investigated.6 There is proven role of TGF beta1 and TGF beta3 in corneal wound healing. TGF beta 1 promotes scarring and beta 3 inhibits it. Use of TGF beta 3 to inhibit the action of TGF beta 1 is being attempted in in-vitro model to prevent the occurrence of sub-epithelial haze following surface ablation. Another treatment still in experimental stage is gene therapy in the form of Targeted AAv5-Smad 7 gene therapy.[28]

Theoretically there is a role of amniotic membrane after PRK ablation to decrease the haze formation as they have anti-inflammatory as well as anti-scarring properties but it seems an impractical solution as post-operative vision will be grossly decreased with use of amniotic membrane. Another modality suggested is trans-epithelial ablation, role of which is yet to be proved.

Surgical treatment should be resorted to if patient is not responding to these modalities and if corneal scar has become permanent in nature. In these cases, superficial debridement with application of topical MMC followed by polishing with burr has been suggested. The role of Phototherapeutic keratectomy (PTK) along with MMC in these cases has been suggested. If nothing works, then sutureless superficial anterior lamellar keratoplasty may be needed, although in very few cases, to restore the visual acuity and clarity of cornea.

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Resistance In Herpes Simplex and Varicella Zoster Viral Infections - A Brief Overview

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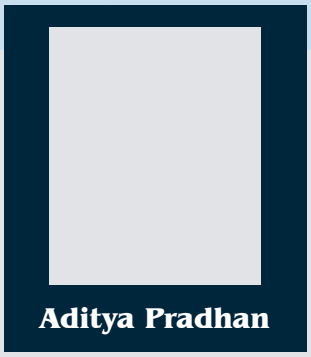

Introduction

Herpes simplex viruses 1 (HSV-1) and 2 (HSV-2) cause orolabial and genital infections as well as keratitis, encephalitis and neonatal infections. Varicella-zoster virus (VZV) is the causative agent of varicella and herpes zoster. Farooq et al have estimated the incidence of HSV keratitis in developed nations to be around **242000 cases/year** and to be **1-1.5 million cases/year** in developing nations[1]. There are no epidemiological studies available from India regarding the same.

Viral DNA polymerase (pol), as well as viral thymidine kinase (TK), is one of the early proteins produced in infection and is responsible for chain elongation. It is the target for antiviral drugs such as acyclovir, ganciclovir, famciclovir, valacyclovir, foscarnet and cidofovir, which all have activity against HSV. **All antiviral agents target the viral DNA pol**[2].

The first-line drugs include deoxy-guanosine analogues such as acyclovir (ACV) and their L-valyl-ester prodrugs valaciclovir (VACV) and famciclovir (FCV). ACV and penciclovir (PCV) must be phosphorylated by the thymidine kinase of HSV or VZV and then by cellular kinases to be converted into their active metabolites. Their triphosphate forms compete with deoxynucleotide triphosphates (dNTPs) for incorporation into the replicating DNA chain. In addition, ACV triphosphate acts as a DNA chain terminator.

The second-line drug is the pyrophosphate analogue foscarnet (FOS) that is administered intravenously for the treatment of nucleoside analogue-resistant HSV and VZV infections. **FOS does not need any activation step to exert its antiviral activity** and thus directly binds to and blocks the pyrophosphate-binding site on the viral DNA pol, which prevents incorporation of incoming dNTPs into viral DNA. Topical and intravenous formulations of cidofovir (CDV) may be used 'off label' in the treatment of nucleoside analogue resistant and/or FOS-resistant HSV infections. CDV is an acyclic nucleoside phosphonate that only requires phosphorylation by cellular enzymes to be activated. Once in its diphosphate form, CDV inhibits the viral DNA pol by acting as a DNA chain terminator.

Emergence of resistance

In immunocompetent individuals, the prevalence of HSV isolates resistant to ACV ranges from 0.1-0.7%3. In immunocompetent patients with herpetic keratitis, the prevalence is around 6.4%4. Resistance is the most in immunocompromised patients at around 3.5-14% and as high as 36% in haematopoietic stem cell transplant recipients[3]. The picture is not so clear as regards to VZV isolates resistant to ACV with only a few reports available.

Mechanisms of resistance

In HSV clinical isolates, resistance to ACV is mediated in 95% of the cases by mutations in the viral TK and, in the remaining cases, by mutations in the viral DNA pol3,5. In VZV clinical isolates, resistance to ACV is mostly conferred by mutations in the viral TK and, less frequently, by mutations in the viral DNA pol[5]. Mutations in TK account for the majority of cases with ACV resistance[4,6]. Patients with ACV resistant strains of HSV caused by a TK mutation are also resistant

to VACV, ganciclovir, and FCV because these antiviral agents also rely on TK phosphorylation. Antiviral agents that do not rely on TK phosphorylation (FOS, CDV, and trifluridine) will not share cross-resistance to acyclovir when there is a TK mutation.

Evaluation of HSV and VZV drug susceptibility

There are phenotypic and genotypic assays available to measure drug susceptibility and resistance. Levels of drug resistance (EC50 values) for HSV are best measured by cell-based (phenotypic) assays. The gold standard phenotypic method to determine the susceptibility of HSV isolates to antiviral drugs is the plaque reduction assay (PRA) in Vero cells that is approved as a standard protocol by the Clinical and Laboratory Standards Institute[7].

Susceptibility of VZV to antiviral drugs can be tested by the PRA using fibroblastic cells[8]. However, the clinical utility of the PRA is limited by the low rate of VZV isolation most exclusively from vesicle fluids and its slow growth in cell culture (typically 5–6 days)[9].

Genotypic assays are too complex to describe in this overview and the interested reader can refer to the excellent review article by Piret et al[A].

Resistance in the Indian scenario

Pramod et al analysed seven HSV-1 isolates from keratitis cases which were clinically resistant to ACV. 4 out of the 7 isolates proved ACV resistant by PRA and 3 of them showed reduced TK activity[10].

Abraham et al undertook a cross-sectional study to determine the ACV susceptibility of HSV- 1 & 2 isolates using a dye uptake assay. 130 isolates from 141 patients were analysed and the overall prevalence was found to be 5.4% (3% in HSV-1 and 7.8% in HSV-2)[11].

Risk factors

Long-term prophylaxis in the form of oral ACV has been implicated as a risk factor for resistance. However, the evidence is conflicting. In a study of isolates from 239 patients who had previously been on suppressive ACV treatment for 6 years, the median ACV sensitivity was not statistically significantly different from that of ACV-naïve patients[12]. A similar study found no significant difference in the incidence of ACV resistance between patients who previously received ACV treatment and treatment-naïve immunocompetent patients (0.67% vs. 0.42%, respectively)[13].

The prevalence of ACV-resistant HSV isolates specifically in ocular infections was assessed in two studies. The first analysed 40 HSV-1 isolates from 35 patients using a dye uptake method and found that one isolate was resistant whereas three had reduced sensitivity. Thirteen of the cases had keratitis[14]. In a second study of 173 immunocompetent patients with HSV keratitis, 11 (6.4%) had ACV-resistant isolates. Ten of the 11 had mutations in the viral TK gene conferring the resistant phenotype. Interestingly, one isolate had cross-resistance to foscarnet as well[4].

Van Velzen et al in an elegant study showed that long-term ACV prophylaxis (≥ 12 months) predisposes to ACV-refractory disease due to the emergence of corneal ACV resistant HSV-1. Recurrence duration of ≥45 days was also identified as an independent risk factor[15].

The high incidence of ACV-resistance in this population of patients can be explained because the cornea can be regarded as an immune-privileged site, with low immune surveillance allowing for rapid selection of mutant viruses. Furthermore, the same ACV-resistant virus could be recovered from the cornea during consecutive herpetic keratitis episodes, indicating that the ACV-resistant HSV-1 is able to establish latency and reactivate[15].

A recent report demonstrated the coexistence of ACV-sensitive and ACV-resistant latent HSV-1 in the trigeminal ganglia from immunocompetent individuals[16].

Role of PCR

Polymerase chain reaction (PCR) is a valuable tool in the diagnosis of HSV/VZV keratitis. A real-time polymerase chain reaction (Real-Time PCR), also known as quantitative polymerase chain reaction (qPCR), is a laboratory technique of molecular biology based on the PCR. It monitors the amplification of a targeted DNA molecule during the PCR, i.e. in real-time, and not at its end, as in conventional PCR. Real-time PCR can be used quantitatively (quantitative real-time PCR), and semi-quantitatively, i.e. above/below a certain amount of DNA molecules (semi quantitative real-time PCR).

Pramod et al studied the utility of PCR analysis

against other diagnostic markers: HSV isolation on cell culture, HSV antigen detection by indirect immunofluorescence, detection of anti-HSV IgG by enzyme-linked immunosorbent assay (ELISA) and detection of HSV-specific tear secretory IgA (sIgA) by ELISA. These tests showed overall sensitivity values of 22.4%, 39.8%, 30.4% and 20.3% respectively[17].

Hlinomazová et al found that the sensitivity of RT-PCR for detection of HSV-1 infection was higher than 90%. They also diagnosed 16 cases of ACV resistant keratitis[18].

Encouraging results regarding the utility of RT-PCR were also reported by Aliabadi et al and Ma et al[19,20].

Kowalski et al reported the results of a new test called AmpliVue that can detect HSV-1/2 DNA and provide results within 1 hour. They found that the sensitivity of AmpliVue against ocular samples that were both culture-positive and PCR-positive was 84%. The specificity of AmpliVue was 100%[21].

Management

The persistence of active lesions due to HSV (not necessarily keratitis) for more than 7–10 days after initiation of high-dose oral ACV, VACV or FCV therapy without apparent decrease in size, an atypical appearance or the emergence of satellite lesions is suggestive of treatment failure. The persistence of clinical signs of VZV infection for more than 10–14 days after initiation of ACV therapy is suggestive of treatment failure. An algorithm is given below[A].

Novel strategies and the road ahead

Some promising compounds are currently in clinical trials. The orally bioavailable lipid ester prodrug of CDV (i.e., hexadecyloxypropyl-cidofovir; brincidofovir) could avoid the dose-limiting toxicity of the parent drug and provide a safe alternative for ACV-resistant herpes viruses in immune-compromised patients[22]. Pritelivir, a potent orally bioavailable helicase primase inhibitor, reduced the rates of genital HSV-2 shedding and days with lesions in a phase II trial[23]. The bicyclic nucleoside analogue FV-100 and carboxylic nucleoside analogue valomaciclovir were well tolerated and effective for the treatment of herpes zoster in phase II trials[24,25]. Novel classes of antiviral agents targeting the ribonucleotide reductase, the helicase-primase complex and the process of viral DNA encapsidation are at earlier stages of development[25].

Conclusion

Resistance of HSV to ACV is a well-established problem which hasn't been given due importance in light of emerging resistance to antibacterials and antifungals. Surveillance programs elsewhere are targeted towards bacteria and don't exist in our country. The treating clinician must have a logical approach towards infectious keratitis. There should be a high index of suspicion in cases which are bilateral, which are immune-compromised, in recurrent HSVK and those showing poor response to treatment. Long term prophylaxis with ACV should be in selected cases and not routinely although it seems inevitable in certain scenarios like post corneal transplantation[B].

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Dosage and formulation of Antibiotics / Antifungals / Antiamoebics

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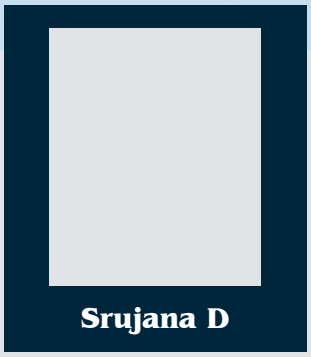
Srujana D

S.No.	Name of antibiotic/ antifungal/ antiamoebics (available dose)	Fortified for topical use: dose & preparation	*Subconjunctival/ #Intracameral: dose & preparation	Intravitreal: dose & preparation
1	Ceftazidime (500mg)	50mg/ml (5%) 1. Add 2 ml of distilled water to 500mg 2. Add to 8ml of artificial tears Refrigerate in 4□C Shelf Life: 1week at 4□C and 3 days at room temperature	*Dose : 100mg/0.5ml	Intravitreal Ceftazidime : 2.25mg/0.1ml 1. Add 2 ml of distilled water to 500mg (250 mg/ml) 2. Take 0.1 ml of solution(25mg/0.1ml) 3. Dilute with 0.9 ml of sterile water(25mg/10ml) 4. Discard 0.9 ml, use 0.1 ml for injection Can be mixed with steroid preparation in the same syringe. Use dexamethasone in Step 3 if indicated
2	Cephazolin (500mg)	50mg/ml(5%) 1. Add 2 ml of distilled water to 500mg 2. Add to 8ml of artificial tears Refrigerate in 4□C Shelf Life: 1week at 4□C and 3 days at room temperature	*Dose : 100mg/0.5ml	
2	Vancomycin (500mg)	50mg/ml(5%) 1. Add 2 ml of distilled water to 500mg 2. Add to 8ml of artificial tears Storage: Refrigerate at 4□C Shelf Life: 28 days at 4□C	*Dose : 25mg/0.5ml	Intravitreal Vancomycin : 1mg/0.1ml 1. Add 5 ml distilled water to 500mg of vancomycin powder for injection 2. Mix well (shake)—100 mg/ml 3. Take 0.1 ml of drug solution 4. Dilute with 0.9 ml of sterile water 5. Mix well (moving air bubble up and down) 6. Discard 0.9 ml 7. Use 0.1 ml for injection Caution: Do not mix with other drugs as it gets precipitated
3	Tobramycin	14mg/ml(1.4%) 1. Add 2ml(80mg) of parenteral tobramycin to commercially available tobramycin eye drops 0.3% 5 ml (15mg/5ml). Shelf Life: 1 week in refrigerator at 4□C and 4 days in room temperature	*Dose : 20mg/0.5ml	

S.No.	Name of antibiotic/ antifungal/ antiamoebics (available dose)	Fortified for topical use: dose & preparation	*Subconjunctival/ #Intracameral: dose & preparation	Intravitreal: dose & preparation
4	Gentamicin	14mg/ml(1.4%) Method: Add 2ml/80mg of parenteral gentamicin to commercial gentamicin ophthalmic solution 0.3% 5 ml (15mg/5ml). Shelf Life: 1 week at 4 degrees C and 4 days in room temperature	*Dose : 20mg/0.5ml	
5	Amikacin	2.5% Method: Parenteral Amikacin 250mg/2ml is mixed with 8 ml artificial tears. Shelf Life: 7 days at 4□C		Intravitreal Amikacin: 125 μg/0.1ml(previously recommended 400 μg was highly retinal toxic) 1. Take 0.1 ml solution (12.5 mg/0.1 ml) of commercially available preparation (250 mg/2 ml) 2. Dilute with 0.9 ml of distilled water (12.5mg/10ml) 3. Discard 0.9 ml,take 0.1 ml solution(1.25mg/0.1ml) 4. Dilute again with 0.9 ml of distilled water(1.25mg/10ml) 5. Use 0.1 ml for injection(125μg/0.1ml)
6	Linezolid 200mg/100ml (2mg/ml)	2 mg/ml (0.2%) Parenteral Linezolid available as IV infusion can be directly used		
7	Colistin (1million IU/75mg)	0.19% Add to 10ml distilled water – 7.5mg/ml (0.75%) 1ml of above solution is then added to 3ml distilled water		
8	Imipenem(500mg)–Cilastin(500mg)	1% Add 10ml sterile water to create a solution of strength 50mg/ml. Take 1 ml of this solution and add 4 ml sterile water to make topical Imipenem 1% - 1mg/ml Storage - In amber coloured bottles Stability – 3 days at 2-8 □C		Intravitreal Imipenem 50– 100 μg/0.1ml 1. Dilute commercially available preparation with (250 mg)100 ml of distilled water 2. Take 0.2 ml (0.5 mg) 3. Dilute with 0.3 ml sterile water 4. Inject 0.05 ml
9	Trimethoprim/ sulfamethoxazole 16 mg/mL and 80 mg/mL	Commercial preparation can be used.		
10	Polymyxin B (500,000 units lyophilized powder)	10,000-25,000 units per mL Dissolve 500,000 polymyxin B units in 20 to 50 mL sterile water for injection or sodium chloride injection USP for a 10,000 to 25,000 units per mL concentration. Note: Avoid total systemic and ophthalmic instillation over 25,000 units/kg/day.	Subconjunctival injection of up to 100,000 units/ day may be used for the treatment of Pseudomonas aeruginosa keratitis	

S.No.	Name of antibiotic/ antifungal/ antiamoebics (available dose)	Fortified for topical use: dose & preparation	*Subconjunctival/ #Intracameral: dose & preparation	Intravitreal: dose & preparation
11	Piperacillin/tazobactam (2g/250mg)/vial: (2.25 g)	10% Add 22ml of 0.9% Sodium Chloride for Injection/ Sterile Water for Injection/ Dextran 6% in Saline/ Dextrose 5% to 2g/250mg Piperacillin/ tazobactam powder to obtain 10% solution Note: Lactated Ringer's solution is not compatible		Intravitreal 225 μg/0.1ml
12	Amphotericin B	0.15% 1. Add 10 ml 5% Dextrose to 50mg of amphotericin B powder. 2. Draw 3 ml of this and add to 7ml of artificial tears eye drops. Storage: Refrigerate in 4□C. Shelf life: 7 days at 4□C and 4 days in room temperature Phototoxic, to be dispensed in brown bottle	#5 -10 μgm/0.1ml For 10μg/0.1ml : 1. Add 10 ml of 5% Dextrose to create 5 mg/ml. 2. Take 0.2ml, dilute with 0.8ml of 5% Dextrose 3. Take 0.1ml, add 0.9ml of 5% Dextrose (0.1mg/1ml) 4. Use 0.1ml(10 μg/0.1ml)	Intravitreal amphotericin B (5μg/0.1 ml) 1. Add 10 ml of 5% Dextrose to commercially available preparation (50mg) and mix well (5 mg/ml) 2. Take 0.1 ml of drug solution (0.5mg/0.1ml) 3. Dilute with 0.9 ml of sterile water (0.5mg/10ml) 4. Again take 0.1 ml of solution (50μg/0.1ml)and add 0.9 ml distilled water (50μg /10ml) 5. Discard 0.9 ml of solution, use 0.1 ml (5μg/0.1ml)
13	Voriconazole (200mg)	1% Mix 20 ml ringer lactate to 200 mg voriconazole lyophilized powder. Stability: 30days at 4□C or room temperature	50 μg/0.1ml (for intrastromal injection) Method: Take 1ml from1% voriconazole solution, add 19 ml ringer lactate to make 0.05mg/ml (50 μgm/0.1ml)	Intravitreal Voriconazole 50– 100 μg/0.1ml 1. Add 19 ml of distilled water to 200 mg voriconazole lyophilized powder (10 mg/ml) 2. Take 0.1 ml solution and add 0.9 ml distilled water and mix well 3. Discard 0.5 ml of solution, take 0.5 ml of sterile water and mix well (air bubble) 4. Discard 0.9 ml of solution, use 0.1 ml for injection (One and a half dilution)
14	Polyhexamethylene biguanide/ Chlorhexidine (20% stock solution)	0.02% 20% stock solution diluted with 0.9% NaCl to obtain 0.02% eye drop solution (0.01ml of stock solution + 10ml of 0.9% normal saline=0.02%)		

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Post LASIK Infective Keratitis

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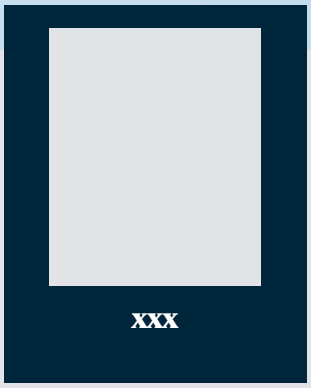

Introduction

The last decade has seen a rising trend in the number of patients opting for laser vision correction (LVC) for refractive error. The various options for laser vision correction include photorefractive keratectomy (PRK), laser subepithelial keratomileusis (LASEK), laser in situ keratomileusis (LASIK) and Small Incision Lenticule Extraction (SMILE). LASIK still remains one the most commonly performed LVC surgery. Complications of LASIK include dry eye, glare, halo, under-correction and over-correction.(1) Post LASIK microbial keratitis is one of the most dreaded sight threatening complication that can result in permanent loss of patients best corrected visual acuity (BCVA).(1) The incidence of post LASIK microbial keratitis reported in literature vary from 0%-1.5%.(2)

Risk Factors

As prevention is better than cure, following risk factors should be kept in mind while assessing every case and avoided as far as possible.(3)

1. **Pre-operative**

   All cases planned for any laser refractive surgery should be screened for the following risk factors

   a. Dry eye disease
   b. Meibomian gland dysfunction
   c. Blepharitis
   d. Immunocompromised status (HIV positive)

2. **Intra-operative**
   a. Breach in OT sterilization or surgical asepsis
   b. Use of same microkeratome blade for multiple cases
   c. Corneal epithelial defect
   d. Presence of interface debris

3. **Post-operative**
   a. Use of a bandage contact lens (BCL)
   b. Splashing of tap water in the eyes
   c. Post traumatic flap dislocation
   d. Poor compliance with medications

Etiology

Post LASIK infective keratitis can be classified into early and late based on its onset from surgery. The microorganisms implicated are different for these two conditions.(2)

1. Early onset infectious keratitis: It is defined as infective keratitis occurring within 2 weeks of surgery. The microorganisms implicated in early onset infective keratitis are primarily gram-positive bacteria like Staphylococcus aureus, Staphylococcus epidermidis, Streptococcus viridians and Streptococcus pneumoniae. Gram negative organisms like Pseudomonas aeruginosa are less commonly associated.(2)
2. Late onset infectious keratitis: It is defined as infective keratitis occurring between 2 weeks to 3 months after surgery. The micro-organisms most commonly implicated in late-onset infective keratitis are fungi, atypical mycobacteria and Nocardia.(2,4) The atypical mycobacteria are present in the environment, soil and body surface, and can readily cause infection as they are highly resistant to chemical disinfection. (5,6)

Clinical Features

Symptoms

The onset of symptoms usually occurs after 2-3 days of surgery. Patient presents with complaints of pain, redness, watering, photophobia, diminution of vision, foreign body sensation and ocular irritation. (2) The symptoms of pain may not be as severe as seen in routine cases of infective keratitis due to severing of the corneal nerves during LASIK.

Signs

On examination, there are corneal infiltrates which is not only limited to the interface but may also involve the flap as well as the deep stromal tissue.(Fig-1, Fig-2) There is inflammatory reaction in the anterior chamber as well.(5) In severe cases, there may be associated flap melting, which heals with corneal scarring resulting in irregular astigmatism. (7)

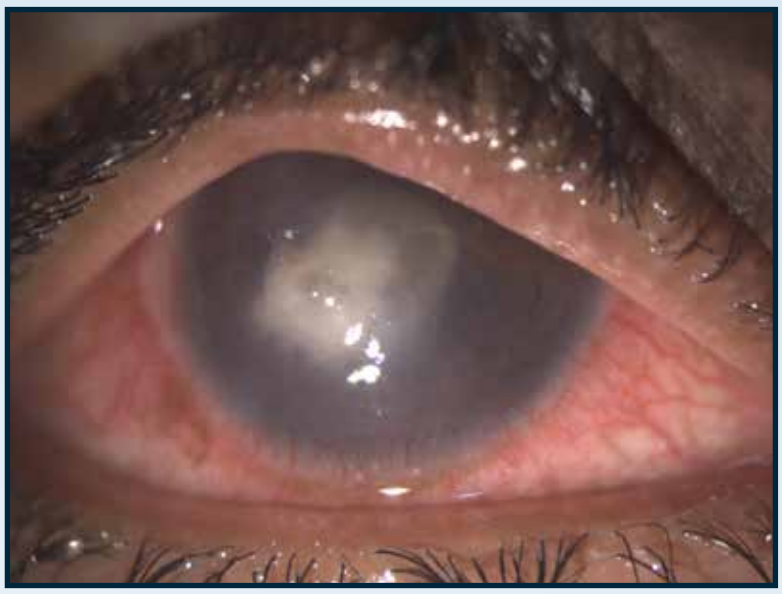

Figure 1. *Slit lamp photograph of right eye showing deep stromal infiltrates with total cataract post LASIK*

Figure 2. *Slit lamp photograph of left eye showing resolving keratitis with stromal infiltrate and surrounding scar post LASIK*

The corneal infiltrate in case of microbial keratitis with gram positive organisms are usually located at the flap edge and have a well defined margin while that due to atypical Mycobacteria and fungi are located at the interface and have an indistinct and feathery margin.(8)

Differential Diagnosis

Other conditions which may be considered as differential diagnosis are(9)

1. Diffuse lamellar keratitis
2. Interface debris
3. Sterile peripheral infiltrates
4. Epithelial in-growth

The most important differential diagnosis for a case of post LASIK infective keratitis is diffuse lamellar keratitis. The differentiating features have been highlighted in Table 1.

Table 1.

Characteristics	Infective Keratitis	Diffuse Lamellar Keratitis
Etiology	Staph. aurous, Staph. epidermidis, Strept. Pneumonia, atypical mycobacteria, Fungi, Nocardia	Inflammatory reaction
Onset	After 2-3 days of surgery	Within first 24 hours
Symptoms	Moderate to severe pain and photophobia	Mild pain and photophobia
Conjunctival congestion	Moderate to severe	Mild
Lid oedema	Moderate to severe	Mild
Location of infiltrate	Interface , may involve the flap and deep stroma	Confined to interface, begins in the flap periphery
Appearance	Focal area of infiltration surrounded by diffuse inflammation	Diffuse interface inflammation; called sands of Sahara
Anterior camber reaction	Present	Absent

Management

The management of Post LASIK keratitis has been comprehensibly described by Donnenfeld et al. (Figure3) (3) In most of the cases there are no superficial corneal infiltrate that can be scraped for sampling. Therefore, it is recommended to lift the flap and scrape the bed for obtaining samples both for smear and culture sensitivity examination.(3) The culture media include blood agar and chocolate agar for bacteria, Sabouraud's dextrose agar for fungus, and Thioglycolate broth for anaerobic organisms. In case of late onset infection, atypical mycobacteria needs to be ruled out and therefore sample is sent in Lowenstein-Jensen or Middlebrook 7H-9 agar in addition to the other culture media. The sample is also subjected to smear examination with Gram stain, Gomori-methenamine silver stain, and Ziehl-Neelsen stain to rule out unusual pathogens such as Nocardia, atypical mycobacteria, and fungi. In cases where all reports are negative, corneal biopsy or polymerase chain reaction is done. In addition, confocal microscopy may be used as an adjunctive non-invasive imaging tool for detection of fungal hyphae, Acanthamoeba trophozoites and cysts.

The treatment of all cases of post LASIK infective keratitis consists of flap lift with scraping of bed for procuring samples for microbiological evaluation. This is followed by irrigation of the flap interface with an antibiotic solution, which consists of fortified vancomycin 5% (50 mg/mL) for early onset cases and fortified amikacin 3.5% (35 mg/mL) for late onset cases of infective keratitis. (3)

Empirically, all cases of early onset infection are started on topical fourth-generation fluoroquinolone (gatifloxacin 0.3% or moxifloxacin 0.5%) along with fortified cefazolin 5% (50 mg/mL) on hourly regimen round the clock for first 48 hours and then 2 hourly regimen until there is clinical response. In case the patient works in a hospital environment and has risk of exposure to methicillin-resistant Staphylococcus aureus (MRSA), topical vancomycin 5% (50 mg/mL) is started instead of cefazolin as it is more effective against MRSA. In addition, oral doxycycline, which has anti-collagenase effect, is given 100 mg BD to prevent further collagenolysis. In case of late-onset keratitis, amikacin 3.5% (35 mg/mL) along with a fourth-generation fluoroquinolone (gatifloxacin 0.3% or moxifloxacin 0.5%) is preferred on hourly regimen. Topical clarithromycin 1%(10mg/Ml) and oral clarithroycin 500 mg BD is also effective and are used in addition topical amikacin 3.5%. (2,6,8) Natamycin 5%(50mg/mL) for filamentous fungi and amphotericin B 0.15% (1.5mg/mL) for yeast infection and topical Voriconazole 1% (10mg/mL) with oral Voriconazole 200 mg BD is effective against both filamentous and yeast infections.(4,10) Topical corticosteroids are discontinued in all these cases. The treatment is further modified based on the culture and sensitivity reports.

In case of progressive keratitis, not responding to medical treatment, partial or total amputation of the flap, based on the location and size of infiltrate is done to allow better penetration of the drug.(11) The healing of infective keratitis results in scar formation and irregular astigmatism. Visual rehabilitation of these cases is later done with either rigid gas permeable contact lens or lamellar/penetrating keratoplasty.

Prophylaxis

The following standard precautions can reduce the risk of post LASIK infective keratitis.(3)

1. Preoperative: Treatment of meibomian gland disease
2. Intraoperative
   a) Proper sterilization of instruments
   b) Separate instruments for both eyes when performing bilateral surgery
   c) Use of sterile drapes, gowns, gloves, and masks by the treating physician and assisting technician
   d) Povidone–iodine solution (Betadine 10%) for lid preparation
   e) Application of drape to remove the lashes from the surgical field
   f) All fluids applied to the eye before, during, and after LASIK should be sterile
3. Post-operative
   a) Topical antibiotic (4^{th} generation floroquinolone)
   b) Use of eye shields in the early post operative period to avoid trauma
   c) Avoid rubbing of eyes
   d) Avoid direct application of tap water in eyes, swimming and gardening
   e) Avoid eye make-up
   f) Frequent lubrication in subjects with dry eye

Conclusion

Post LASIK infective keratitis is a dreaded complication of refractive surgery. Proper pre-operative, intra-operative and post-operative precautions can avert this sight threatening complication. In addition, an early diagnosis with timely initiation of appropriate treatment (medical/surgical) is necessary for best outcome.

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Riboflavin & Types of Corneal Collagen Crosslinking

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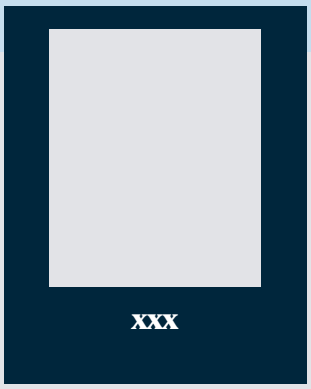

Introduction

Keratoconus (KC) is an asymmetric, progressive corneal ectasia that can leads to significant visual impairment due to associated myopia and irregular astigmatism. The gold standard management of progressive KC has been corneal collagen crosslinking (CXL), using ultraviolet-A (UVA) radiation and the photosensitizer riboflavin. This procedure has the potential to stop the progression of keratoconus, avoiding the need for penetrating keratoplasty.(1)

Keratoconus progression varies with age with most rapid progression is seen in puberty, which continues throughout adolescence, and stabilizes at around third or fourth decade of life. This stabilization has been explained by the age-related increase in corneal stiffness which is due to the glycation induced cross-linking between or within stromal collagen lamellae, as a result of the accumulation of non-enzymatic glycation end products over time.(1)

Corneal collagen crosslinking (CXL)

CXL was introduced in 1995, as a technique to increase the mechanical stiffness of connective tissue. (2,3) In this technique 0.1% aqueous solution of riboflavin 5 phosphate (vitamin B2) acts as a photomediator and absorbs the UV A radiation (370 nm) to generate free radicals thereby, inducing the formation of additional intra-molecular collagen crosslinks. This molecule also provides adequate shielding of the deeper tissues such as the endothelium, lens and macula. This technique has been shown to arrest progression of keratoconus due to increase in biomechanical strength of the human cornea by approximately 300%.(3)

The riboflavin solution is diluted in dextran 20%, a carrier to make it iso-osmolar to the corneal stroma in order to avoid any stromal swelling. The corneal epithelium acts as a barrier against the penetration of this macromolecule (riboflavin) which necessitates the debridement of epithelium prior to instillation of riboflavin.(4)

Mechanism

When activated with UVA light, riboflavin is converted into excited singlet and triplet states, which undergo a series of chemical reactions subsequently, by two mechanisms. A mechanism favored at low oxygen concentration (Type I), produces radical riboflavin, while a mechanism favored at higher oxygen concentrations (Type II), produces singlet molecular oxygen. The radical riboflavin and reactive oxygen species produced by the Type I & II photochemical mechanisms are thought to interact with corneal proteins (such as tyrosine and tryptophan) to create chemical bonds that result in corneal cross-linking. Therefore, the presence of riboflavin in the cornea both protects the internal structures of the eye from UVA transmittance and induces the cross-linking effect.(5)

The Dresden (standard) protocol

This standard protocol uses iso-osmolar 0.1% riboflavin solution, which is generated by diluting vitamin B2-riboflavin-5-phosphate 0.5% with

dextran T500 20% (402.7 mOsmol/L). After a 9 mm diameter debridement of the corneal epithelium, iso- osmolar riboflavin solution is applied on the cornea every 3 minutes for 30 minutes. Following this, ultrasound pachymetry (five repetitive measurements) should be performed at the thinnest point to ensure a minimal stromal thickness of 400 μm.(3,6) Successful penetration of riboflavin is ensured by visualization of riboflavin in the anterior chamber on slit-lamp biomicroscopy. The eye is then irradiated for 30 minutes with UV-A at an intensity of 3 mW/cm2, corresponding to a surface dose of 5.4 J/cm2 at a working distance of 5 cm (UV-X; IROC Medical, Zurich, Switzerland; distributed by Peschke Meditrade GmbH, Nuremberg, Germany) to induce corneal crosslinks to a depth of around 310 μm. An isotonic 0.1% riboflavin solution and a topical anesthetic (oxybuprocaine 0.4% or 0.5% proparacaine) are administered every 5 minutes to saturate the cornea with riboflavin.

This protocol has been reported to be efficacious in slowing or halting the progression of ectasia in patients with keratoconus, with a significant improvement in maximum keratometry (Kmax) or cone apex power in CXL-treated eyes relative to controls at 12 months, postoperatively.(7,8) A commonly used riboflavin is the Peschke® D standard riboflavin (0.1%) solution with 20% dextran 500.

Modifications for thin corneas

According to the current inclusion criteria a minimal stromal thickness (after debridement of the corneal epithelium) of 400 μm is required for performing CXL in order to avoid endothelial damage. However, to treat the patients with thin corneas various modifications have been introduced to induce stromal swelling and increase the stromal thickness prior to CXL.

Hypo-osmolar Riboflavin (0.1%)

Hypo-osmolar 0.1% riboflavin solution is formed by diluting vitamin B2-riboflavin-5-phosphate 0.5% with physiologic salt solution (sodium chloride 0.9% solution; 310 mOsmol/L; B. Braun Medical AG, Sempach, Switzerland) instead of dextran. The procedure performed is similar as described above that is after removal of the corneal epithelium and 30 minutes of instillation of iso-osmolar riboflavin solution, the corneal stromal thickness is measured using ultrasound pachymetry. In cases where the thickness is less than than 400 μm, hypo-osmolar riboflavin is applied every 20 seconds for 5 additional minutes, and the corneal thickness is again measured following which the administration of riboflavin is continued till the thickness reaches 400 μm.

The absolute increase in the corneal thickness reported in a case series ranged from 36 to 110 μm, where the thinnest cornea was 323 μm after removal of the epithelium. The results of this technique are noted to be similar to those of the standard protocol (ie, an isoosmolar solution).(9)

Different types of ribloflavin commonly used are:

- Peschke® D: Standard riboflavin solution for epi-off procedure containing 0.1 % Riboflavin (Vitamin B2) with 20% Dextran (Dresden original)
- Peschke® M: Standard riboflavin solution without dextran for epi-off procedure containing 0.1 % Riboflavin (Vitamin B2) with 1.1 % HPMC.
- Peschke® H: a hypotonic riboflavin solution for inducing corneal swelling containing 0.1 % Riboflavin (Vitamin B2)

Transepithelial Corneal Collagen Cross-linking

Riboflavin is known not to readily penetrate the intact epithelium, this has led to the development of different techniques to increase the absorption of riboflavin into the stroma, including the use of eye drops containing preservatives, such as benzalkonium chloride (BAK) preoperatively. (10) The efficacy of transepithelial CXL has been reported to be only one fifth of the conventional CXL with complete epithelial removal. (11) The main advantages of this technique include improved patient comfort and decreased risk of infection associated with epithelial removal.

Riboflavin with BAK has been shown to effectively penetrate the intact epithelium. BAK acts as a tensioactive substance and changes the surface tension value, facilitating the penetration of other substances through biological membranes. It has been shown in experimental studies that with this technique, the epithelial surface retains its curvature due to increased stiffness with more compact arrangement and straightening of the stromal fibres. This effect was noticed in the upper 50% of the stroma. Thus, tensioactive CXL is a less invasive

procedure with fewer complications and increased patient comfort. (12)

One of the examples of riboflavin used for this technique is Peschke® TE which is recommended to be instilled for a duration of 20 minutes (1 drop every 2 minutes = 10 drops) and contains 0.25 % Riboflavin (Vitamin B2) with 1.2 % HPMC and 0.01 % Benzalkoniumchloride.

Riboflavin with ethylenediaminetetraacetic acid (EDTA)

Filippello et al has shown the demarcation line to be more superficial (100 μm) as compared to that in conventional CXL (320 to 340 μm) using 0.1% aqueous riboflavin involving trometamol and sodium EDTA to break down epithelial intercellular junctions. (13) This may suggest a more superficial effect of transepithelial CXL.

Other modifications

Customized pachymetry-guided epithelial debridement

This technique includes cone-on CXL that is preservation of the epithelium over the cone (thinnest area of cornea) or area of maximal topographical steepening. [14] However, this technique has been shown to have a limited penetration of riboflavin with a demarcation line reported to be at 150 μm.(15)

Contact lens assisted CXL (CLCXL)

Riboflavin-soaked bandage contact lens has been used effectively in thin corneas for CXL and is reported to augment the corneal thickness by 100 μm approximately. [16] However, the major limitations include inability to customize the contact lens thickness and intraoperative buckling. Moreover, the absorption properties of contact lenses may be different from that of the corneal stroma. Long-term results of this procedure are yet not available.

Refractive lenticule assisted CXL

Augmentation of stromal thickness for CXL in thin corneas has also been achieved using refractive lenticules obtained from patients undergoing small incision lenticule extraction (SMILE) for myopic correction.[17] This technique is believed to be the most physiological method to increase the stromal thickness. Moreover, the rough host stromal surface allows the lenticule to spread easily and buckling is avoided.

Hydroxypropyl methylcellulose (HPMC) Riboflavin

The use of hydroxypropyl methylcellulose riboflavin has been shown to prevent corneal dehydration induced by dextran and is more suitable for thin corneas. [18] Riboflavin solution with 20% dextran leads to approximately 30% corneal thinning, whereas a hydroxypropyl methylcellulose (HPMC)–containing solution resulted in less than 10% corneal thinning and a hypotonic saline solution resulted in 10% corneal swelling. (19)

Adapted fluence

This technique is evolving as the latest technique for thin corneas and is based on the principle of customizing UV-A irradiance according to the stromal thickness. The UV-A fluence delivered in all the previously described techniques is constant (5.4 J/cm2) whereas, this technique involves delivering of customized energy by altering the irradiation time (UVA irradiation of 3 mW/cm2 for a customized irradiation time).(20) This also eliminates the need for varying riboflavin concentrations and overcomes the limitations associated with other techniques.

Iontophoresis (I-CXL)

Iontophoresis is a noninvasive technique that involves application of a mild electric current and allows effective penetration of transepithelial riboflavin. Since, riboflavin is a negatively charged, water-soluble molecule with a relatively low molecular weight, it is suitable for iontophoresis.(21,22) Initial studies have demonstrated stabilization of the disease process following iontophoresis-assisted CXL (I-CXL).(23) However, the keratometric regression was lower as compared to conventional epi-off cross-linking.(24) Also the depth of penetration and concentration achieved is noted to be deeper and greater as compared to transepithelial CXL but still less than epi off conventional method.(25) Thus, I-CXL has the potential to become an alternative treatment for keratoconus while reducing the treatment time, postoperative patient discomfort as well as risk of infection.

Accelarated CXL

Accelerated CXL aims at reducing the duration of the procedure and improving the patient comfort. These protocols are based on the fundamental law of photochemistry, called as the Bunsen-Roscoe Law of Reciprocity. (26,27) This law states

that photochemical biological effect of ultraviolet is proportional to the total energy dose delivered, regardless of the applied irradiance and time. This law holds true within the range of irradiances of currently available UVA delivery devices. The energy dose delivered by a UVA source (joules per square centimeter) is the product of the irradiance of the source (watts per square centimeter) and the delivery time (seconds).

However, several studies have evaluated the safety and efficacy of accelerated protocols in the range of 7 to 45 mW/cm2 irradiation with a similar efficacy and visual outcome without any significant endothelial cell loss.(26,28-30) Thus, the Bunsen and Roscoe reciprocity law is valid only for illumination intensity up to 45 mW/cm2 and an irradiance time >2 min. However, the demarcation line is reported to be more pronounced and more anteriorly located than the standard protocol.(30-32) Thus, a shallow demarcation line in accelerated CXL also indicates reduced risk of endothelial cytotoxicity. (33)

The major limitation reported with this protocol is the reduced corneal stiffening effect with increasing intensity of UV-A radiation at more than 18 mW/cm2. This is mainly due to the rapid oxygen consumption and failure of Type I photochemical reaction at an intensity of more than 10 mW/cm2 thereby leading to reduced efficacy and higher failure rates.(34)

Pulsed illumination

Pulsed illumination during CXL is based on the concept that illumination with UVA irradiation causes a rapid depletion of oxygen in cornea saturated with riboflavin, while turning the UVA lamp off leads to rapid replenishment of the oxygen to its original level by diffusion from the tissues.(12) This further acts to create additional cross-linking interactions. (35,36) This technique using 0.1% riboflavin with HPMC (VibeX Rapid; Avedro) with a UVA dose of 7.2 J/cm2 with a pulse interval of 1 seconds has been reported to have a deeper demarcation line (200 μm) as compared to 160 μm in continuous group. No endothelial cell loss was observed in either group. (37)

However, further studies are required to determine the ideal pulsing duration and approach.

Customized cross linking / Photorefractive intrastromal CXL (PiXL)

Various authors have studied the factors predicting the outcome of CXL. The only factors independently predictive of improvement after CXL were CDVA of 20/40 or worse or Kmax of 55.0 D or greater. (38, 39) There was no role of corneal thickness, preoperative corneal haze, and cone location in predicting the outcome of CXL. However, a more eccentric cone has also been shown to be associated with a higher keratometry at 1year follow-up. (40)

This technique involves delivering of customized UVA treatment patterns localized on specific corneal zones. This pattern is generated using a three-dimensional finite element analysis (FEA) model. (41) The KXL II; Avedro device incorporating iris tracking and programmable UVA patterns has recently been granted CE Mark in Europe and is being used to study customized CXL (referred to as photorefractive intrastromal CXL [PiXL]). The UVA LED source illuminates a digital micromirror device (DMD). This technique has shown a significant improvement in visual acuity in keratoconus as well as in patients with hyperopia.(42,43)

Laser in situ keratomileusis Xtra (LASIK Xtra)

This technique is based on the concept of compensating the reduced biomechanical strength of the cornea in LASIK. However, the major concern is the flattening effect resulting in hyperopic shift as well as the post-CXL stromal haze, which can contribute to deterioration in visual quality. [44]

The LASIK Xtra procedure involves application of a higher concentration (0.25%) riboflavin on the stromal bed subsequent to excimer laser ablation for a duration of 90 s followed by washing of interface and repositioning of the flap. UV-A irradiance is then delivered as a homogenous beam of 30 mW/cm2 for 90 s (2.7J/cm2). This delivers around half the energy used in conventional cross- linking in the Dresden protocol, which helps to restore or improve corneal strength without inducing a refractive change.

Various studies have shown a greater refractive accuracy and stability with a lower incidence of regression in hyperopes with this procedure. (45-47)

One of the examples of riboflavin used for this technique is Peschke® L which contains > 0.23 % Riboflavin (Vitamin B2).

Conclusion

CXL with riboflavin and UVA irradiation is a minimal invasive technique that modifies corneal stroma and increases the biomechanical stability. It is still an evolving technology and has the potential to maximize the visual rehabilitation with various upcoming modifications in the technique.

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