Update on diagnosis and management of refractory corneal infections

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Infectious keratitis is a global cause of concern for visual disability and corneal blindness. A refractory corneal ulcer can be defined as an ulcer with an inadequate healing response to conventional therapy. Scant evidence exists in the literature on a clear clinical definition or specified time duration of refractory keratitis. Under normal conditions, once the infective component is neutralized, corneal ulcers heal due to the proliferative ability of the corneal epithelium. However, various systemic, ocular, and organism characteristics predispose to the development of non-healing or refractory corneal ulcers. Prompt etiological diagnosis and appropriate antimicrobial therapy constitute the mainstay of treating infectious corneal ulcers; however, an inadequate response results in progressive worsening requiring surgical intervention, leading to a poor outcome in refractory keratitis.

This review explores the current literature for various factors contributing to infective keratitis refractory, diagnosing, and managing them along with probing future directions. In this article, we have considered ulcers with inadequate healing response to conventional treatment, ulcers worsening on treatment, and refractory or virulent organisms or infiltrates in specific post-surgical interventions such as laser refractive surgery (LRS), keratoplasty, or post collagen cross-linking based on their location and the altered local tissue response as refractory keratitis.

Contributory factors

Factors, both ocular and systemic, can contribute to a refractory ulcer [Table 1]. Identifying them will not only help in early diagnosis and timely management but also help prevent ulcers from becoming refractory.

Systemic factors

Systemic risk factors such as diabetes, use of oral steroids/immunosuppressives, and underlying autoimmune conditions weaken the ocular immune system, increasing the severity of the infection and resulting in inadequate or delayed response to treatment. Lim et al. noted them to be significant risk factors for polymicrobial keratitis as compared to monomicrobial infections. Diabetes is an independent risk factor for fungal infection, which correlates with the severity of the infection and worsens the prognosis. Similarly, the use of systemic immunosuppressive medications is known to exacerbate the severity and delay fungal clearance. O’Neill et al. reported diabetes, systemic immunosuppression, and use of systemic steroids/oral immunosuppressives to be independent risk factors for microbial keratitis-associated endophthalmitis, thus making it necessary to manage the immunosuppressed state effectively and, if possible, to discontinue the immunosuppressives for a while after consulting with the treating physician/rheumatologist.

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Table 1: Ocular and systemic factors contributing to a refractory ulcer

| Ocular Factors          | Systemic Factors                          |
|-------------------------|-------------------------------------------|
| Ulcer profile           | Uncontrolled diabetes mellitus            |
| Inaccurate Diagnosis    | On oral Immunosuppression                 |
| Coexisting ocular diseases | Immunosuppressed state                  |
| Superadded infection    | Malnutrition                              |
| Antimicrobial resistance|                                          |
| Refractory organism     |                                          |
| Specific situations (Post PK/LK/LRS/CXL) |                          |

PK - penetrating keratoplasty, LK - lamellar keratoplasty, LRS - laser refractive surgery, CXL - collagen cross-linking

Ocular factors

Ulcer profile

Signs of a healing ulcer include decreased symptoms, reduced ulcer size, infiltrates and hypopyony, epithelialization, and finally, scarring. However, cases refractory to conventional medical therapy show worsening of most of the abovementioned features in addition to progressive corneal stromal melt/thinning.[2,3]

Recurrence of infection, indolent ulcers, neurotrophic ulcers, ulcers larger than 6 mm in size, deep stromal or full-thickness infiltrates, and impending perforation are clinical profiles of refractory keratitis apart from those caused by multidrug-resistant or virulent organisms such as Pythium insidiosum or Pseudomonas aeruginosa or a polymicrobial infection. Indolent slow-growing infiltrates tend to resist susceptibility to potent antimicrobials due to biofilm formation.[8] Deep stromal infiltrates and endothelial plaques in chronic mycotic ulcers have poor penetration and accessibility of therapeutic agents to the depth of posterior stroma and endothelium, with the overlying epithelium having healed. This leads to recalcitrant fungal infections,[8] necessitating the need to adopt a targeted therapeutic approach for drug delivery.[8,9]

Diagnosis

An ulcer more than 2 mm in size or involving the visual axis must be investigated microbiologically.[9] An inaccurate diagnosis or empirical therapy with multiple medications causes surface toxicity and alters the clinical picture in addition to leading to an inadequate response to treatment, thus emphasizing the need for microbial tests.

Basic diagnostic techniques

The mainstay in the diagnosis of corneal ulcers is an examination of corneal smears obtained by corneal scraping and culture of corneal samples.[9-11] Gram stain accurately detects causative organisms 60%–75% of the time for bacterial cases[12] and 35%–50% for fungal.[13] Potassium hydroxide (KOH) wet mount has a sensitivity of 76.3% for diagnosing fungal keratitis.[8,13] Calcofluor white stain is helpful in fungal, Acanthamoeba, and Microsporidal keratitis.[13,14] Blood and chocolate agar are the most commonly used culture media for bacteria. Sabouraud’s dextrose agar or potato dextrose agar is best for isolating fungi, and non-nutrient agar enriched with Escherichia Coli is employed to culture Acanthamoeba.[13-15]

Clinically refractory fungal keratitis can also be reviewed for Pythium insidiosum. The hyphae of P. insidiosum stain positive for calcofluor-KOH, acridine orange hydrochloride, and lactophenol blue,[14] and it grows well in blood, Sabouraud’s dextrose, and chocolate agar,[16] however, polymerase chain reaction (PCR) is considered as the diagnostic test.[16]

Viral keratitis is primarily a clinical diagnosis.[8,9] PCR is noted to be highly sensitive, especially in diagnosing various viral pathogens such as herpes simplex virus, adenovirus, and cytomegalovirus, along with multiple other organisms such as bacteria, fungus, Acanthamoeba, and microsporidiosis.[27]

In a retrospective study of 23897 cases of presumed keratitis over 10 years at Aravind Eye Hospital in India, 38% of corneal scrapings tested negative, both on culture and smear.[18] Culture-negative keratitis remains a significant problem for clinicians in the management of refractory keratitis. If the ulcer is refractive to empirical therapy and cultures are negative, repeat cultures of the ulcer and referral to a cornea specialist may be warranted. While doing repeat scraping, it is recommended that antimicrobial therapy be stopped at least 24–48 h prior.[19] When the repeat culture of a progressive, non-responding corneal ulcer is negative, histological examination of the corneal biopsy specimen is indicated. Superficial keratectomy or corneal biopsy specimen can be obtained by a trephine or free lamellar dissection with a sharp blade for immunohistochemical and light-microscopic examination. This approach is beneficial for the detection of fungi and Acanthamoeba in deep ulcers.[20] Despite repeating these basic investigations not infrequently the organism remains unidentified and there arises a need to look for alternate/advanced options.

Advanced diagnostic techniques

Internal transcribed spacer (ITS) gene sequencing establishes a rapid and prompt diagnosis of fungal keratitis in refractory cases[21] and has been described in Pythium along with other non-sporulating molds.[22-33]

Molecular identification also helps diagnose rare fungal species such as Beauveria bassiana, which was found to be highly resistant to antifungal therapy, along with Colletotrichum gloeosporioides and Trametesbetulin.[22,34,35]

Apart from these, in vivo confocal microscopy (IVCM), a non-invasive method, is increasingly being used due to its rapidity and high sensitivity in detecting larger and deep-seated organisms inaccessible by routine scraping, such as filamentous fungus, Acanthamoeba, and Nocardia.[18,40][Fig. 1]. Anterior segment optical coherence tomography (AS-OCT) has been used to provide an objective measure of the size of the corneal infiltrate/scar dimensions or to monitor the progress of corneal thinning during treatment.[41,42]

Next-generation sequencing (NGS) has emerged as a novel method that may improve the diagnostic accuracy of infectious keratitis, particularly for organisms that are difficult to culture by conventional methods such as atypical or anaerobic bacteria.[43] However, it is not clear whether these approaches can be used to effectively determine the etiology of infection or antibiotic sensitivity data.[44]
Emerging New Pathogenic Microbes

Several new pathogenic fungi causing keratitis, with varying or suboptimal susceptibility to antifungal therapy, are emerging [Table 2]. Knowledge of the sensitivity profile of antifungals by antifungal susceptibility test (AFST) against various species helps in initiating appropriate treatment and improving the outcome. A recent study from South America reported A. fumigatus isolate from post-traumatic keratitis in a 27-year-old male worker carrying the substitution G54E at Cyp51Ap associated with itraconazole resistance, highlighting the possibility of mutation-induced resistance to common antifungal therapy.

Coexisting Ocular Diseases

The ocular surface is directly exposed to the environment, where it interacts with a myriad of pathogens. The gel-forming mucins and tight intercellular junctions of the epithelium prevent the entry of organisms, and tears help flush the noxious substances out of the eye, help maintain healthy epithelium, and limit the growth of pathogens with the help of proteins such as lysozyme, immunoglobulins, and lactoferrin. If any of the abovementioned factors or mechanisms are overwhelmed...
Table 2: Details of emerging new fungal corneal infections reported in recent literature

| Study                                                                 | Risk                        | Microorganism                      | AFST                                                                 |
|----------------------------------------------------------------------|-----------------------------|------------------------------------|----------------------------------------------------------------------|
| [23]Tan SJ et al. Contact lens associated keratitis due to Tintelnotiadestructans. Med Mycol CaseRep. 2019; 27: 8-10. | CL wear Immuno-compromised CL wear | Tintelnotiadestructans               | amphotericin B, ciclopirox, natamycin, posaconazole, voriconazole, and terbinafine |
| [24]Kaufmann et al. Tintelnotiadestructans Keratitis: A Clinicopathological Report and Review of the Literature. Cornea. 2021; 40: 380-382. | Trauma                     | Lasiodiplodiapseudotheobromae (dematiaceous fungi) | voriconazole and amphotericin B                                      |
| [25]Behrens-Baumann WJ et al. Keratomycosis due to Tintelnotiadestructans refractory to common therapy treated successfully with systemic and local terbinafine in combination with polyhexamethylene biguanide. IntOphthalmol. 2019; 39: 1379-1385. | Trauma                     | Aspergillus tamarii                  | Azoles                                                                |
| [26]VanamHP et al. First report of Lasiodiplodiapseudotheobromae keratitis susceptible to voriconazole in an Indian mango grower. Access Microbiol. 2019; 1: e000055. | Trauma                     | Pseudallescheria boydii              | voriconazole and posaconazole                                        |
| [27]Homa M et al. Characterization of Aspergillus tamarii Strains from Human Keratomycoses: Molecular Identification, Antifungal Susceptibility Patterns and Cyclopiazonic Acid Producing Abilities. Front Microbiol. 2019; 10: 2249. | Trauma                     | Aspergillus viridinutans             | Micafungin                                                            |
| [28]Shigeysasu C et al. Keratomycosis caused by Aspergillus viridinutans: an Aspergillus fumigatus-ressembling mold presenting distinct clinical and antifungal susceptibility patterns. Med Mycol. 2012; 50: 525-8. | Trauma                     | Purpureocillium lilacinum            | voriconazole                                                         |
| [29]Rosa PD et al. Antifungal Susceptibility, Morphological and Molecular Characterization of Lasiodipodiatheobromae Isolated from a Patient with Keratitis. Case Reports Mycopathologia. 2018; 183: 565-571. | Trauma                     | Curvularia. tainlandensis, Curvulariacoimbatorensis | natamycin and amphotericin B                                          |
| [30]Kiss N et al. New Species of the Genus Curvularia: C. tainlandensis and C. coimbatorensis from Fungal Keratitis Cases in South India. Pathogens. 2019; 9: 9. | Trauma                     | Curvularia senegalensis              | amphotericin B, miconazole, itraconazole and ketoconazole           |
| [31]Guarro J et al. Mycotic keratitis due to Curvularia senegalensis and in vitro antifungal susceptibilities of Curvularia spp. J ClinMicrobiol. 1999; 37: 4170-3. | Trauma                     | Fusarium keratoplasticum, Fusarium falciforme, Fusarium sporotrichoides | natacamycin and amphotericin B                                       |
| [32]Al-Hatmi AMS et al. Keratitis by Fusarium temperatum, a novel opportunist. BMC Infect Dis. 2014; 14: 588. | Trauma                     | Fusarium temperatum (Fusarium fujikuroi species complex) | micafungin, posaconazole and amphotericin B                          |
| [33]Sun S et al. Identification and Characterization of Fusarium proliferatum, a New Species of Fungi that Cause Fungal Keratitis. Sci Rep. 2018; 8: 4859. | Trauma                     | Fusarium proliferatum               | natamycin and voriconazole                                           |

Contd...
Table 2: Contd...

| Study                                      | Risk                      | Microorganism                        | AFST                  |
|--------------------------------------------|---------------------------|--------------------------------------|-----------------------|
| [59‑63] Monden Y et al. First case of fungal keratitis caused by Pestalotiopsisclavispora. ClinOphthalmo. 2013;7:2261-4 | Multiple ocular surgeries, herpetic infection, bullous keratopathy | Pestalotiopsisclavispora | Micafungin            |
| [57] Gajar DU et al. Severe pigmented keratitis caused by Cladorrhinumbulbillosum. Indian J Med Microbiol. 2011; 29: 434-7. | Immuno-compromised        | Cladorrhinumbulbillosum              | naftamycin, amphotericin B, fluconazole and itraconazole |
| [54] Wang L et al. Fungal keratitis caused by a rare pathogen, Colletotrichumgleosporioides, in an east coast city of China. Case Reports J Mycol Med. 2020; 30: 100922. | Trauma, Topical steroids   | Colletotrichumgleosporioides (filamentous fungi) | amphotericin B, voriconazole, itraconazole, posaconazole, micafungin and caspofungin |
| [51] Hardin JS et al. Fungal Keratitis Secondary to Trametesbetulina: A Case Report and Review of Literature. Mycopathologia. 2017; 182: 755-759. | Trauma                    | Trametesbetulina (filamentous fungi) | voriconazole          |
| [60] Aggarwal S et al. Exophialaphaeomuriformis Fungal Keratitis: Case Report and In Vivo Confocal Microscopy Findings. Case Reports Eye Contact Lens. 2017; 43: e4-e6. | Post PKP                  | Exophialaphaeomuriformis (pigmented yeast) | voriconazole          |

either because of the underlying disease or an adverse effect of treatment, an organism can gain entry. [55] Green et al. [56] identified ocular surface disease (OSD) as a predisposing factor for microbial keratitis. These patients presented with more severe infections, higher incidence of polymicrobial or mixed infections, and took longer to heal. Among the OSDs, blepharitis followed by dry eye, SJS, and OCP were the most prevalent; coagulase-negative Staphylococcus aureus was the most common species with non-healing epithelial defect, resulting in corneal perforation being the most common complication in a five-year study on microbial keratitis with OSD in Australia. [57] Lacrimal duct obstruction or chronic dacryocystitis by delaying the tear clearance alters the ocular flora, thus making the cornea more susceptible to infections. Staphylococcus species is reported to be the most common; however, fungal infections have also been reported. [58] Several measures can be employed to prevent further deterioration of the surface. These include the use of preservative-free drops, tarsorrhaphy, and punctal occlusion in cases of neurotrophic or severe dry eyes to control factors causing underlying inflammation. In addition, a heightened awareness regarding the possibility of altered microbial flora and anticipating delayed epithelialization are essential. A low threshold is adopted for applying cyanoacrylate glue to prevent perforation as stromal melt tends to progress quickly in these compromised eyes. Judicious use of the abovementioned measures aids in faster resolution of non-healing corneal ulcers. [59‑63]

**Superadded infections**

Occasionally, an ulcer with a good healing response could worsen. This indicates either compromised compliance or a superadded/secondary infection. Patient compliance needs to be reaffirmed, and a repeat corneal scraping helps rule out a secondary infection. The presence of an epithelial defect, history of steroid use, and previous recurrent episodes of keratouveitis were identified as risk factors for secondary bacterial and fungal infection in herpes simplex keratitis. [64]

**Specific situations**

a) Post-penetrating keratoplasty

Infected keratitis following optical keratoplasty is one of the important causes of graft failure and poor visual outcome. The predisposing risk factors are grouped into three categories: donor-related (infected donor tissue), host-related (ocular surface disorders, use of topical steroids or contact lens, recurrence of previous infection, or underlying systemic disease), and graft-related (suture related, persistent epithelial defect, or wound leak/dehiscence). [60] Most studies report a higher incidence of infection within the first year of surgery, thus warranting a close follow-up, particularly in those with underlying risk factors stated above. [61] To prevent this, proper surveillance of the donor tissue, and in particular, consideration of intraoperative suturing techniques and wound integrity are essential. Improving the ocular surface health by punctal occlusion, tarsorrhaphy, lid corrective surgeries, and epilation are additional procedures that are planned as required. [62] In addition, oral acyclovir 400 mg twice a day is recommended as a prophylactic dose in patients undergoing a graft for healed viral keratitis; however, the exact duration for which it needs to be continued is unclear. [68] Use of prophylactic antibiotics in the absence of a persisting defect is not recommended as it has limited or no role. [69] Medical management alone with topical/systemic antimicrobials is found to control when the infiltrate is <4 mm in 66% of cases, [62] whereas larger ones require a regraft if the organism is not very sensitive. Despite the resolution of infection, the visual prognosis is poor because of a high incidence of graft failure [Fig. 2]. [65‑67,70]

b) Post-lamellar keratoplasty

The graft host interface remains a potential space for infection to occur following lamellar keratoplasty, and though rare, results in significant visual morbidity. As the site of infection is deep within the stroma, it restricts access to the infiltrate for microbiological testing. In addition, it impacts penetration of topical drugs, thus delaying the diagnosis and response to treatment besides the use
of topical steroids in the postoperative period being a risk factor.\textsuperscript{[71]} Candida species has been the most common organism reported to cause interface infection, followed by Klebsiella.\textsuperscript{[71]} Infected donor tissue was the most common risk factor identified.\textsuperscript{[71],[73]} Tissue warming during the tissue processing for lamellar keratoplasty promoted Candida growth in donor rims. However, the addition of antifungal agents to storage media raised concerns about endothelial toxicity.\textsuperscript{[74,75]} A single or multiple whitish infiltrate/s seen in the interface should raise suspicion of an interface infection warranting close observation. This is especially important because these infections rarely produce significant symptoms. As the infection is deep-seated restricting access to the microbiological sample, confocal microscopy offers additional value; however, the role of anterior segment OCT is limited.\textsuperscript{[76,77]} Based on the donor corneal rim culture and clinical appearance, empirical treatment is initiated with topical and systemic antimicrobials. Washing the interface with antimicrobial agents or deep intrastromal/intracameral injections with antifungal shave has been attempted with limited success, with most cases requiring a therapeutic penetrating keratoplasty.\textsuperscript{[71,78]} Removal of the donor lenticule with the aim of reducing the microbial load too has achieved limited success. This has, on the contrary, led to recurrence of infection in the interface, and of dissemination of infection in the anterior and posterior chamber, causing endophthalmitis in posterior lamellar keratoplasty.\textsuperscript{[79,80]} High degree of clinical suspicion, close watch, especially in eyes with positive donor rim culture reports, along with antimicrobial injections in the interface, should be attempted to avoid further interventions.

c) Post-refractive surgery

The incidence of post-laser refractive surgery (LRS) infection is 0.0001%–1.5%, and it is higher after photorefractive keratectomy (PRK) than laser in situ keratomileusis (LASIK) or small-incision lenticule extraction (SMILE), probably due to a large epithelial defect following PRK.\textsuperscript{[81]} Risk factors include preexisting dry eyes, blepharitis, Meibomian gland dysfunction, intraoperative contamination of instruments or surgical field, and the use of bandage contact lens postoperatively.\textsuperscript{[81,82]} Based on the onset, keratitis is defined as early (within 1 week of surgery) and is usually caused by Staphylococcus/Streptococcus or late (beyond 1 week of surgery) wherein slow-growing organisms such as fungus, mycobacteria, Nocardia, or Acanthamoeba should be suspected. Herpetic keratitis too can present following laser refractive procedure as a primary infection or due to reactivation.\textsuperscript{[82,83]} In flap procedures, the infiltrate usually occurs in the interface or is limited only to the lamellar flap, flap margin, or stroma.\textsuperscript{[82,84]} For microbiological assessment in procedures with an interface, the flap needs to be lifted. The undersurface of the flap or the interface is scraped, followed by a thorough wash with fortified antibiotics. Most cases respond to medical management, but in non-responding cases, repeated interface irrigation, flap amputation, PACK CXL, tissue adhesives, and surgical intervention might be needed.\textsuperscript{[82-84]} Improving the health of the ocular surface preoperatively and a close watch in the postoperative period with timely intervention taking into account the possible microorganism based on their presentation can help improve outcomes [Table 3].

d) Post collagen cross-linking

Infective keratitis post collagen cross-linking (C3R) is rare and most commonly involves staphylococcus species. Large epithelial defect, damage to stromal keratocytes by UV light, use of topical steroids and bandage contact lens postoperatively, and an altered ocular surface in patients with vernal or atopic keratoconjunctivitis or blepharitis might be the predisposing factors and need to be considered.\textsuperscript{[85-87]} Reactivation of the
herpes simplex virus by UV light has been hypothesized by Kymonis et al,[88] to be responsible for causing herpetic epithelial keratitis and uveitis inpatients following C3R. Though most cases respond well to topical antibiotics, it must be borne in mind that in the immediate postoperative period, in the absence of keratocytes, corneal melt in the presence of an infection can proceed very rapidly, at times necessitating a therapeutic penetrating keratoplasty in these eyes [Fig. 3]. A close watch for resistant microorganisms and altered sensitivity patterns should be monitored in case of worsening of infections.

**Management of refractory ulcers**

Antimicrobial therapy forms the mainstay of treatment. However, with the emergence of antimicrobial resistance (AMR), virulent and new pathogens, attention is focused on the development of novel antimicrobial compounds with better penetration and adjunct therapeutic modalities to prevent the need for surgical intervention and augment the treatment response.

**Antimicrobials**

For bacterial keratitis, single-drug therapy using fluoroquinolone has been traditionally the mainstay of management.[88] Combined fortified topical antibiotics should be considered for large and/or visually significant corneal ulcers, especially if a hypopyon is present and for eyes unresponsive to initial treatment.[88] In various studies, including some randomized controlled trials, both moxifloxacin and gatifloxacin performed at least as well as standard fortified cefazolin/tobramycin combination therapy.[88‑90] However, Methicillin-resistant *S. aureus* isolates are generally resistant to fluoroquinolones but susceptible to vancomycin.[91,94] Vancomycin-resistant *S. aureus* is very rare but sensitivity to topical linezolid has been demonstrated in such cases.[95] Keratitis from multidrug-resistant *Pseudomonas aeruginosa* has also been reported, with high morbidity further highlighting the need for antibacterial sensitivity.[93‑96] Topical colistin 0.19%, imipenem, or polymyxin B 10000–20000 IU/ml may be considered in such cases.[95] Systemic antibiotics may be considered in severe cases where the infectious process has extended to adjacent tissues (e.g., the sclera) or when there is impending or frank perforation of the cornea.[88] Systemic therapy is also necessary in cases of gonococcal keratitis.[97] Gram-positive rods (non-tuberculous mycobacteria) can be treated with amikacin, clarithromycin, or azithromycin therapy, whereas gram-positive rods (*Nocardia*) are susceptible to sulfacetamide, amikacin, or trimethoprim/sulfamethoxazole therapy [Table 4].[96]

The use of adjuvant corticosteroids has long been debated in the treatment of bacterial keratitis.[98‑100] Steroids for Corneal Ulcers Trial (SCUT) compared adjunctive topical corticosteroids to placebo in treating bacterial corneal ulcers. Despite the comprehensive data showing no difference in outcomes such as 3-month visual acuity, scar size, or perforation rate, subgroup analyses suggested that corticosteroids are beneficial in specific subgroups.[101]

### Table 3: Infective keratitis associated with kerato-refractive surgical procedures

| Refractive surgery | Site of infection | Organism (most common) | Treatment Recommended |
|--------------------|------------------|------------------------|-----------------------|
| PRK                | Base/edge of epithelial defect | Staphylococci/Streptococci | Topical antibiotics based on antimicrobial sensitivity |
| LASIK              | Flap/interface | Early -Staphylococci/Streptococci | Topical antibiotics based on antimicrobial sensitivity |
| SMILE              | Interface | Late- Candida/Nocardia/Mycobacteria Staphylococci | Interface wash with antibiotics/PACK-CXL |

PRK - Photorefractive keratectomy, LASIK - Laser in-situ keratomileusis, SMILE - Small-incision lenticule extraction

### Table 4: Antimicrobial therapy recommended against various microorganisms causing infective keratitis

| Microorganism | Recommended antimicrobial agents |
|---------------|----------------------------------|
| Gram-positive cocci[63,64,71] | Cefazolin, Vancomycin, Fluoroquinolones, Bacitracin |
| Gram-negative bacilli[63,64,71] | Tobramycin, Gentamicin, Ceftazidime, Fluoroquinolones |
| Gram-negative cocci[63,64,71] | Ceftriaxone, Ceftazidime, Fluoroquinolones |
| Gram-positive bacilli (Non-tuberculous mycobacteria)[71] | Amikacin, Clarithromycin, Azithromycin, Fluoroquinolones |
| Gram-positive bacilli (*Nocardia*)[71] | Sulfacetamide, Amikacin, Trimethoprim, Sulfamethoxazole |
| Methicillin-resistant *S. aureus* (MRSA)[89,92] | Vancomycin |
| Vancomycin-resistant *S. aureus* (VRSA)[70] | Linezolid |
| *Pseudomonas aeruginosa*[40] | Polymyxin B, Colistin |
| Filamentous fungi[4,77,85] | Natamycin, Ketoconazole |
| Yeasts (e.g., *Candida spp.*)[4,77,85] | Amphotericin B, Natamycin, Ketoconazole, Flucytosine |
| Newer/resistant fungal strains[53‑25] | Voriconazole, Posaconazole, Micafungin, Caspofungin, Itraconazole, Fluconazole, Ciclopirox, Terbinafine |
| *Herpes Simplex Virus*[86‑90] | Trifluridine, Acyclovir, Ganciclovir, Valacyclovir |
| *Varicella Zoster Virus*[97,90] | Acyclovir, Ganciclovir, Valacyclovir |
| *Acanthamoeba spp.*[91] | Chlorhexidine, Polymyxamethylene biguanide, Propamidine |
| *Pythium insidiosum*[25,123] | Linezolid, Azithromycin, Topical ethanol |
| *Microsporidium spp.*[94] | Propamidine, Fumagillin, Fluoroquinolones, Albendazole, Itraconazole |

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For fungal keratitis, treatment with topical natamycin 5% is the mainstay of management. Topical amphoterin B 0.15%–0.5% is an alternative primarily for yeasts, but its use requires access to a compounding pharmacy and is limited by toxicity. Voriconazole, a newer generation triazole, has gained popularity in treating fungal keratitis due to its excellent ocular penetration. The first Mycotic Ulcer Treatment Trial (MUTT I) showed a benefit of natamycin over voriconazole for topical treatment of fungal keratitis, particularly for Fusarium keratitis, which was also confirmed by a second randomized clinical trial and a recent Cochrane review. The Mycotic Ulcer Treatment Trial II (MUTT II) investigated the effect of adjuvant oral voriconazole versus oral placebo for smear-positive filamentous fungal keratitis and did not report a significant benefit of adding systemic voriconazole. Therefore, currently, topical natamycin remains the most evidence-based treatment for filamentous fungal keratitis, and oral voriconazole can be considered if the organism is Fusarium, or if there is the risk of impending/frank perforation or associated scleritis. Other potential adjuvant treatments for endothelial plaques in fungal keratitis include intracameral injection of amphotericin or voriconazole with or without hypopyon drainage or intrastromal injection of voriconazole in cases of deep stromal infiltrates [Fig. 4]. Terbinafine has been suggested to be efficacious in treating severe cases of fungal keratitis due to the rare fungi, Tintinblatia destructans, which is refractory to common antifungal therapy. New strains identified within the same mycotic family might exhibit differences in their susceptibility to antifungal agents. Inaccurate etiological diagnosis or ineffective antimicrobial therapy with partially sensitive or resistant therapeutic agents in the setting of empirical antifungal therapy without AFST is responsible for the progression of the ulceration in refractory cases; therefore, AFST is recommended despite the increased economic burden, especially in refractory cases. Resistance to amphotericin B has been found to correlate with the proteinase production ability of filamentous fungi; however, multidrug resistance to antifungal treatment is considered rare.

Management of viral keratitis includes antiviral medications with or without adjuvant topical corticosteroids. Topical acyclovir is the first-line treatment for HSV epithelial keratitis and oral for stromal and endothelial keratitis. Ganciclovir is a newer synthetic medication with more broad-spectrum antiviral coverage. In addition to treating HSV and VZV keratitis, topical ganciclovir is also effective in treating keratitis caused by CMV. Ganciclovir has been shown to be just as effective as acyclovir and can especially be used in patients resistant or intolerant to acyclovir. The Herpetic Eye Disease Study I (HEDS I) evaluated the effectiveness of corticosteroids in treating HSV stromal keratitis. Time to resolution of infection was significantly shorter in the group receiving topical corticosteroid than those taking placebo. Oral valacyclovir, a newer antiviral, is well-tolerated, and there is some evidence that it may have better ocular penetration. Additionally, the treatment dose for valacyclovir is 1 g three times daily, as opposed to acyclovir which is 400 mg five times daily (800 mg five times daily for VZV), which aids in patient compliance. HEDS II examined the prolonged use of oral acyclovir for prophylaxis of recurrent ocular HSV and reported that ocular HSV recurrence was 45% lower in the acyclovir group at 12 months.

Medical therapy for Acanthamoeba keratitis typically begins with topical chlorhexidine 0.02% or a combination of chlorhexidine/polyhexamethylene biguanide 0.02% and propamidine 0.1%. Therapy needs to be continued for 6–12 months. Corticosteroids need to be used with extreme caution only once the infective aspect is well taken care of and are indicated only in cases where the immune component is contributing like uveitis, scleritis, or optic neuritis.

For Pythium insidiosum, various studies have evaluated the efficacy of a combination of topical linezolid with topical and oral azithromycin and have found mixed results. However, most cases are not amenable to medical therapy and early surgical treatment with or without adjuncts may be warranted. Recently, the safety and efficacy profile of topical ethanol in the treatment of Pythium keratitis was reported; however, the exact dose and strength of ethanol that will be most effective needs further work.

The most appropriate treatment for microsporidial stromal keratitis has not yet been established, and therapeutic keratoplasty is recommended in the majority. Treatment with 0.02% polyhexamethylene biguanide does not offer any significant advantage over placebo. Microsporidial infections in HIV-infected individuals may respond to the combination of antibiotics and antiparasitic agents, including topical propamidine, topical fumagillin, topical fluoroquinolones, oral albendazole, and/or oral itraconazole.

Future perspectives

Biofilm promotes adherence of microbes to the surface, interferes with drug penetration, and increases the resistance to antimicrobials; thus, the need for increased understanding of the role of biofilm formation in infections may aid in the development of improved antimicrobial strategies. The biofilm formation that occurs in Fusarium solani has been cited for functioning as a survival strategy that provides antifungal resistance. The description of efflux pumps in Fusarium solani species complex (FSSC) biofilms and promethazine challenged biofilms showing increased sensitivity to amphotericin B offer prospects to explore this therapeutic strategy for effective management of fusarium infections.

Modifications of the antimicrobials to improve their penetration and efficacy have gained significant importance in recent times. Cyclodextrins are natural cyclic oligosaccharides with a hydrophilic outer surface comprising (α-1,4-) linked α-D-glucopyranose units and a lipophilic central cavity. Hydroxypropyl β CD, a cyclodextrin used as a carrier for ketoconazole, led to a 20-fold increase in drug bioavailability compared to suspensions.

Nanoparticles are the colloidal carriers and can be divided into microcapsules, wherein the drug is generally enclosed in a polymer shell or nanosphere, wherein the drug is uniformly distributed within the polymer. Chitosan oligosaccharides (CS) are naturally biocompatible mucodhesive positively charged polymers. Ofloxacin loaded on CS-modified nanolipid carriers were found to have excellent penetration, improved preocular residence time-controlled drug release, and improved corneal bioavailability.

Liposomes are yet another form of nanoformulations comprising lipid vesicles. Investigators have studied liposomes...
to deliver idoxuridine, fusidic acid, amphotericin B, and minocycline.[130‑133]

Over the past few decades, contact lenses have gained attention to be used as a tool for delivering therapeutics against diseases prevailing in the anterior segment of the eye, including keratitis. Desirable drug-eluting contacts allow the devices to be used for the treatment of eye infections. The mechanism of action by which drug-eluting contact lenses work is through the release of drugs directly to the infected area, providing a sustained release of medication for a longer period of time.

Adjunct measures
Despite maximum antimicrobial therapy, the infective keratitis worsens frequently, resulting in melts, sclera extension, perforations, and endophthalmitis, and, in some cases, panophthalmitis. To circumvent such sight-threatening complications in cases of non-responding corneal ulcers, adjunct measures or alternate therapies can be considered in addition to the continuing treatment modality.[137]

a) Photodynamic therapy (PDT)
PDT involves a non-toxic dye (photosensitizer), a low-intensity visible light (red to the near red range), which in the presence of oxygen combines to produce cytotoxic reactive oxygen species. The two basic mechanisms by which PDT induces lethal damage on the microorganisms are by damaging the DNA and cytoplasmic membrane, thus allowing leakage of cellular contents or inactivation of membrane transport systems and enzymes.[138,139] It is also found to increase the stiffness of the corneal tissue, reduce enzymatic digestion by pathogenic microorganisms, and prevent corneal melt.[140] Most PDT studies have attempted collagen cross-linking by using riboflavin and UVA following the Dresden protocol and termed it a photoactivated chromophore for infectious keratitis (PACK-CXL).[141] Based on available evidence, PACK-CXL is most effective in resolving bacterial keratitis with limited success in fungal keratitis. However, the data is insufficient to comment on acanthamoeba, viral, and mixed infective keratitis.[142,143]

A fundamental difference has been noted in susceptibility to PDT between different organisms because of the variation in their cell membranes and cellular organelles.[138,139] This made researchers to experiment with different permutations of photosensitizers and light, for example, toluidine blue O with red light for bacterial keratitis, methylene blue with argon laser for Candida, and rose bengal (RB) with a green light for Fusarium, Aspergillus, Candida, Acanthamoeba, and methicillin-resistant Staphylococcus aureus (MRSA).[139,140‑145] RB-PDT was reported to have better efficacy than riboflavin CXL in inhibiting fungal growth in an in vitro study.[146] However, the depth of penetration of photosensitizers in inflamed corneas, exact duration/dose, and the possibility of intraocular complications because of the light remains to be determined.

b) Phototherapy
The major disadvantage of PDT is the two-part combination approach (photosensitizer + light), with challenges in introducing the same photosensitizer in different microorganisms along with limitations in tissue penetration of the light. To counter this, only light-based therapy is being investigated as an option. Blue light has been gaining attention due to its intrinsic antimicrobial effect and is supposedly less damaging to mammalian cells than ultraviolet light.[147] The exact mechanism of action is still unclear, but the accepted hypothesis is that it excites endogenous intracellular porphyrins, which produce highly cytotoxic reactive oxygen species, mainly singlet oxygen, similar to PDT.[148‑150]

Lasers produce a coherent, monochromatic, and high-energy form of light, causing photocagulation of the tissue. Argon laser was first used by Fromer et al.[151] to treat Pseudomonas keratitis in rabbit corneas. It causes heating and denaturation, leading to cell death. The temperature of the corneal tissue rises over 90° after argon laser, which is believed to contribute to its fungicidal action and increase the epithelial permeability of antimicrobials.[152,153]

Though heartening results, light therapy still needs further studies to determine appropriate timing, dose, and protocol.

c) Cold plasma
Plasma is an ionized gas and consists of ultraviolet light, electromagnetic fields, visible light, ions, heat radiation, and excited species. The effect of all these single components together leads to the disinfecting effect of plasma.[154] Argon and helium are two gases that have been used for plasma generation.[154,155] Reitberger et al.[154] devised an argon cold plasma
Figure 6: Flowchart depicting a stepwise multidimensional approach for managing refractory corneal ulcers
plasma pen and reported its successful use in treating corneal infections with both bacterial and fungal organisms, in vitro and in vivo. However, the response of microorganisms differs from the cold plasma, thus indicating that factors such as the type of plasma, the distance between the jet and the treating surface, duration of treatment, and characteristic features of microorganisms influence the response to treatment.[125]

d) Alcohol

Alcohols have broad-spectrum antimicrobial activity against almost all microorganisms such as bacteria, fungi, viruses, and Acanthamoeba. The antimicrobial property is optimal between 60% and 90%.[127] It acts mainly on the membrane, alters the pH, increases membrane leakage and inhibits growth, reduces sugar uptake, and increases thermal sensitivity.[128] Agarwal et al.[123] reported the efficacy and safety of topical absolute ethanol in the treatment of Pythium insidiosum keratitis. They noted that the absence of ergosterol in the cell wall of Pythium makes it more susceptible to ethanol as compared to fungi in in vitro studies. The authors recommend an outpatient procedure for placing a cotton swab soaked in absolute (99.9%) ethanol over the corneal infiltrate for 60 s, in the supine position following application of topical anesthesia and an eye speculum. Repeat applications are based on the improving clinical response. Ethanol is also found to have cytotoxic effects on Acanthamoeba cysts in addition to the trophozoites, and pretreatment with ethanol was found to be safe and effective in controlling Acanthamoeba keratitis in 20 of 24 eyes.[129,130] However, further studies are required to determine the exact dose and duration of treatment.

e) Cryotherapy

Cryotherapy has been used in the treatment of herpes virus and pseudomonas keratitis.[131,132] The possible mechanism of the efficacy include mechanical destruction of microorganisms because of intra and extracellular ice formation, osmotic disequilibrium, disruption of DNA, and other cellular and enzymatic changes.[104] The cooling effect is reported to be best when the diameter of the freezing point is 1 mm larger than the freezing head, with freezing temperatures ranging from −50°C to −60°C for a freezing time of 6–7 s.[105] Cryotherapy causes denaturation and degradation of proteins within fungal cell walls, resulting in their fracture.[104] These cryotherapeutic actions have been found to be cidal for the trophozoites but not for the acanthamoeba cysts.[105] The cornea can tolerate freezing till the endothelium is intact and can regenerate after freeze injury helping regain the normal transparency.[106,107]

Surgical management

Relentless worsening of the infiltrate, limbal involvement, impending or actual perforations, are indications for urgent surgical or specialist measures.

a) Worsening infiltrate

Despite all measures, in eyes with worsening of the infiltrate, extension to and beyond the limbus, a therapeutic graft is recommended to avoid spillover to and involvement of adjacent tissues leading to sclerokeratitis or endophthalmitis. Recurrence has been noted in the graft host interface post lamellar keratoplasty; however, in cases with the stromal participation alone, a deep anterior lamellar keratoplasty can be attempted with a thorough wash of the interface with antimicrobials intraoperatively.[108,109] In full-thickness infiltrates, a penetrating keratoplasty with graft size 0.5–1 mm larger than the clinically involved area is recommended as there is always a potential risk of subclinical persistence of organisms near the edges of the lesion, and a postoperative histopathological examination helps confirm whether the disease clearance was adequate. A preoperative ultrasound B-scan is advised to rule out the possibility of endophthalmitis.[172] The use of cryotherapy and ethanol as surgical adjuncts in infiltrates involving the limbus has been recommended to address macroscopically invisible involvement, thus reducing the possibility of recurrence. Single freeze-thaw cryotherapy at the trephined edge mark prior to entering the anterior chamber, followed by application of 99.9% ethanol using a sponge placed for 60 s, intraoperatively seemed to reduce the need for a repeat graft and helped salvage the globe in patients with Pythium keratitis [Fig. 5].[173]

b) Impending/small perforations

In cases with progressive corneal stromal melt or descemetocele, tissue adhesives such as cyanoacrylate glue provide the much-required tectonic support and reduce the possibility of perforation.[174] In addition, they also have bacteriostatic action and induce vascularization to promote healing. The use of cyanoacrylate glue is successful in sealing perforations up to 2–3 mm, thus avoiding the need for a therapeutic graft.[172] A conjunctival flap advocated by Gunderson, by bringing in blood vessels to the infected area, facilitates faster healing and provides tectonic support, and can be considered as a treatment option.[173] Halim et al. compared the results of amniotic membrane (AMT) and conjunctival flap in eyes with refractory non-viral infectious keratitis with impending/small perforations. They found both to be effective in providing metabolic and mechanical support for corneal healing in accordance with other published reports.[174–176] AMT acts as a biological bandage that promotes epithelialization and may have an antimicrobial effect.[177]

c) Large perforations

Perforations larger than 3 mm usually require an urgent full-thickness patch graft or a penetrating keratoplasty as irreversible angle closure and secondary glaucoma or expulsive hemorrhage may occur if the anterior chamber remains flat or the eye remains hypotonic.[170,177] AMT and amniotic membrane (AMT) are shown to help stabilize the anterior chamber and provide redundancy for the involved area. These grafts are effective in repairing perforations of up to 5 mm, with good long-term outcomes;[178] however, AMT cannot be used in the presence of coagulative necrosis.[179] Halim et al.[173] reported its successful use in treating corneal perforations. They have also been shown to increase the epithelialization and corneal clarity in eyes with central keratitis.[171] The authors recommend an outpatient procedure for placing a patch graft or a penetrating keratoplasty as discussed above.[171,177–179]

Conclusion

To conclude, managing refractory corneal ulcers necessitates a stepwise multidimensional approach, involving early and accurate diagnosis, identification of associated factors contributing to its non-responsive behavior (whether local or systemic), and addressing each of them [Fig. 6]. With advances in recent research, newer modalities of treatment have been shown to supplement or act as an alternative therapy for resistant microbial keratitis and improve the overall prognosis.

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Conflicts of interest

There are no conflicts of interest.
References

1. Ting DSJ, Ho CS, Deshmukh R, Said DG, Dua HS. Infectious keratitis: An update on epidemiology, causative microorganisms, risk factors, and antimicrobial resistance. Eye (Lond) 2021;35:1084-101.
2. Ziaei M, Greene C, Green CR. Wound healing in the eye: Therapeutic prospects. Adv Drug Deliv Rev 2018;126:162-76.
3. Katzman LR, Jeng BH. Management strategies for persistent epithelial defects of the cornea. Saudi J Ophthalmol 2014;28:168-72.
4. Lim NC, Lim DK, Ray M. Polymeric versus monomicrobial keratitis: A retrospective comparative study. Eye Contact Lents 2013;39:348-54.
5. Dan J, Zhou Q, Zhai H, Cheng J, Wan L, Ge C, et al. Clinical analysis of fungal keratitis in patients with and without diabetes. PLoS One 2018;13:e0196741.
6. Wu TG, Keasler VV, Mitchell BM, Wilhelmus KR. Immunosuppression affects the severity of experimental Fusarium solani keratitis. J Infect Dis 2004;190:192-8.
7. O’Neill EC, Yeoh J, Fabini DC, Cassidy D, Vajpayee RB, Allen P, et al. Risk factors, microbial profiles and diagnosis of microbial keratitis-associated endophthalmitis in high-risk eyes. Graefes Arch Clin Exp Ophthalmol 2014;252:147-62.
8. Agrawal V, Biswas J, Madhavan H N, Mangat G, Reddy MK, Saini JS, et al. Current perspectives in infectious keratitis. Indian J Ophthalmol 1994;42:171-92.
9. Mootha V, Shahinpoor P, Sutton DA, Xin L, Najafzadeh MJ, Hoog GS. Identification problems with sterile fungi, illustrated by a keratitis due to a non-sporulating Chaetomium-like species. Case Reports Med Mycol 2012;50:361-7.
10. McLeod SD, Kolahdouz-Isfahani A, Rostamian K, Flowers CW, Lee PP, McDonnell PJ. The role of smears, cultures, and antibiotic sensitivity testing in the management of suspected infectious keratitis. Ophthalmic Epidemiol 1996;10:23-28.
11. Kim E, Chidambaram JD, Srinivasan M, Lalitha P, Wee D, Lietman TM, et al. Prospective comparison of microbial culture and polymerase chain reaction in the diagnosis of corneal ulcer. Am J Ophthalmol 2008;146:714-23, 723 e1.
12. Chang HY, Chodosh J. Diagnostic and therapeutic considerations in fungal keratitis. Int Ophthalmol Clin 2011;51:33-42.
13. Badiee P, Nejabat M, Alborzi A, Keshavarz F, Shakiba E. Comparative study of Gram stain, potassium hydroxide smear, culture and nested PCR in the diagnosis of fungal keratitis. Ophthalmic Res 2010;44:251-6.
14. Zhang W, Yang H, Jiang L, Han L, Wang L. Use of potassium hydroxide, Giemsa and calcofluor white staining techniques in the microscopic evaluation of corneal scrapings for diagnosis of fungal keratitis. J Int Med Res 2010;38:1961-1967.
15. Gopinathan U, Sharma S, Garg P, Rao GN. Review of epidemiological features, microbiological diagnosis and treatment outcome of microbial keratitis: Experience of over a decade. Indian J Ophthalmol 2009;57:273-9.
16. Grooters AM, Whittington A, Lopez MK, Boroughs MN, Roy AF. Evaluation of microbial culture techniques for the isolation of Pythium insidiosum from equine tissues. J Vet Diag Invest 2002;14:288-94.
17. El-Aal AM, El Sayed M, Mohammed E, Ahmed M, Fathy M. Evaluation of herpes simplex detection in corneal scrapings by three molecular methods. Curr Microbiol 2006;52:379-82.
18. Lalitha P, Prjna NV, Manoharan G, Srinivasan M, Mascarinas J, Das M, et al. Trends in bacterial and fungal keratitis in South India, 2002-2012. Br J Ophthalmol 2015;99:192-4.
19. Lin A, Rhee MK, Akpek EK, Amescua G, Farid M, Garcia-Ferrer FJ, et al. Bacterial keratitis preferred practice pattern (R). Ophthalmology 2019;126:1-P 75-5.
20. Lee P, Green WR. Corneal biopsy: Indications, techniques and a report of a series of 87 cases. Ophthalmology 1990;97:718-2.
21. Kumar M, Shukla PK. Use of PCR targeting of internal transcribed spacer regions and single-stranded conformation polymorphism analysis of sequence variation in different regions of rRNA genes in fungi for rapid diagnosis of mycotic keratitis. J Clin Microbiol 2005;43:662-8.
22. Figueira L, Pinheiro D, Moreira R, Pinto E, Simões J, Camisa E, et al. Beauveria bassiana keratitis in bullous keratopathy: Antifungal sensitivity testing and management. Eur J Ophthalmol 2012;22:814-8.
23. Tan SJ, Nure M, Gardam D, McKnight C, Boan PA, Clark BM. Contact lens associated keratitis due to Tintelnotia destructans. Med Mycol Case Rep 2019;27:8-10.
24. Vanam HP, Ather M, Madhura KS, Rudramurthy SM. First report of Lasiodiplodia pseudothelueomorae keratitis susceptible to voriconazole in an Indian mango grower. Access Microbiol 2019;1:e000055.
40. Vaddavalli PK, Garg P, Sharma S, Sangwan VS, Rao GN, Thomas R. Role of confocal microscopy in the diagnosis of fungal and acanthamoeba keratitis. Ophthalmology 2011;118:29-35.

41. Konstantopouls A, Kuo J, Anderson D, Hossain P. Assessment of the use of anterior segment optical coherence tomography in microbial keratitis. Am J Ophthalmol 2008;146:534-42.

42. Martone G, Pichierrri P, Franceschini R, Moramarco A, Ciompì L, Tosi GM, et al. In vivo confocal microscopy and anterior segment optical coherence tomography in a case of alternaria keratitis. Cornea 2011;30:449-53.

43. Papatioannou L, Miligkos M, Papathanassiou M. Corneal collagen cross-linking for infectious keratitis: A systematic review and meta-analysis. Cornea 2016;35:62-71.

44. Iseli HP, Thiel MA, Hafezi F, Kampmeier J, Seiler T. Ultraviolet A/riboflavin corneal cross-linking for infectious keratitis associated with corneal melts. Cornea 2008;27:590-4.

45. Kaufmann C, Arnold M, Schipf A, Bruderer VL, Iselin KC. Tintennotia destructans keratitis: A clinicopathological report and review of the literature. Cornea 2021;40:380-2.

46. Behrens-Baumann WJ, Hofmüller W, Tammer I, Tintelnot K. Keratomyocysis due to tintennotia destructans refractory to common therapy treated successfully with systemic and local terbinafine in combination with polyhexamethylene biguanide. Int Ophthalmol 2019;39:1379-85.

47. Chew R, Dorma A, Woods ML. Purpuraeocilium limacium keratitis: A case series and review of the literature. Can J Ophthalmol 2016;51:382-5.

48. Todokoro D, Yamada N, Fukuchi M, Kishi S. Topical voriconazole therapy of Purpuraeocilium limacium keratitis that occurred in disposable soft contact lens wearers. Int Ophthalmol 2014;34:1159-63.

49. Guarro J, Akiti T, Horta RA, Morizot-Leite- Filho LA, Gené J, Ferreira-Gomes S, et al. Mycotic keratitis due to Curvularia senegalensis and in vitro antifungal susceptibilities of Curvularia spp. J Clin Microbiol 1999;37:4170-3.

50. Sreepurna AT, Al-Hatmi AMS, Kindo AJ, Sundaram M, Hoog GS. Multidrug-resistant Fusarium inkeratitis: A clinic-no mycological study of keratitis infections in Chennai, India. Mycoses 2017;60:230-3.

51. Gajjar DU, Pal AK, Santos JM, Ghodadra BK, Vasavada AR. Severe pigmented keratitis caused by Cladophorinibulum. Indian J Med Microbiol 2011;29:434-8.

52. Aggarwal S, Yamaguchi T, Dana R, Hamrah P. Exophiala phaeomuriformis Fungal Keratitis: Case report and in vitro confocal microscopy findings. Eye Contact Lens 2017;43:e4-6.

53. Leonardelli F, Theill L, Nardin ME, Macedo D, Dudiuk C, Mendez E, et al. First irtraconazole resistant aspergillus fumigatus clinical isolate harbouring a G54E substitution in Cyp51Ap in South America. Rev Iberoam Micol 2017;34:46-48.

54. Mantelli F, Mauris J, Argüeso P. The ocular surface epithelial barrier and other mechanisms of mucosal protection: From allergy to infectious diseases. Curr Opin Allergy Clin Immunol 2013;13:563-8.

55. Narayanan S, Redfern RL, Miller WL, Nichols KK, McDermott AM. Dry eye disease and microbial keratitis: Is there a connection? Ocul Surf 2013;11:75-92.

56. Green M, Apel A, Stapleton F. Risk factors and causative organisms in microbial keratitis. Cornea 2008;27:22-7.

57. Khoo P, Cabrera-Aguas M, Robaei D, Lahra MM, Watson S. Microbial keratitis and ocular surface disease: A 5-year study of the microbiology, risk factors and clinical outcomes in Sydney, Australia. Curr Eye Res 2019;44:1195-202.

58. Li G, Guo J, Liu R, Hu W, Xu L, Wang J, et al. Lacrimal duct occlusion is associated with infectious keratitis. Int J Med Sci 2016;13:800-5.

59. Kang BS, Kim MK, Woo WR, Oh JY. Infectious keratitis in limbal stem cell deficiency: Stevens-Johnson syndrome versus chemical burn. Cornea 2016;35:51-5.

60. Bagga B, Motukupally SR, Mohamed A. Microbial keratitis in Stevens-Johnson syndrome: Clinical and microbiological profile. Ocul Surf 2018;16:454-7.

61. Dua HS, Said DG, Messmer EM, Rolando M, Benitez-Del-Castillo JM, Hassain PN, et al. Neurotrophic keratopathy. Prog Retin Eye Res 2018;66:107-31.

62. Iyer G, Srinivasan B, Agarwal S. Ocular sequelae of Stevens-Johnson syndrome: A comprehensive approach. Cornea 2020;39:S3-6.

63. Larrañaga Fragoso P, Boto de Los Bueis A, Bravo Ljubetic L, Del Hierro Zarruvelo A, Romero Gómez MP, Mora Rillo M. Herpes simplex keratitis in rheumatoid arthritis patients. Ocul Immunol Inflamm 2016;24:282-7.

64. Boisjoly HM, Pavan-Langston D, Kenyon KR, Baker AS. Superinfections in herpes simplex keratitis. Am J Ophthalmol 1983;96:354-61.

65. Vaippeey RB, Sharma N, Sinha R, Agarwal T, Singhvi A. Infectious keratitis following keratoplasty. Surv Ophthalmol 2007;52:1-12.

66. Akova YA, Onat M, Koc F, Nurozluer A, Duman S. Microbial keratitis following penetrating keratoplasty. Ophthalmic Surg Lasers 1999;30:449-55.

67. Davila JR, Mian SI. Infectious keratitis after keratoplasty. Curr Opin Ophthalmol 2016;27:358-66.

68. Tambasco FP, Cohen EJ, Nguyen LH, Rapuano CJ, Laibson PR. Oral acyclovir after penetrating keratoplasty for herpes simplex keratitis. Arch Ophthalmol 1999;117:445-9.

69. Tavakkoli H, Sugar J. Microbial keratitis following penetrating keratoplasty. Ophthalmic Surg 1994;25:356-60.

70. Harris DJ Jr, Stulting RD, Waring GO 3rd, Wilson LA. Late bacterial and fungal keratitis after corneal transplantation. Spectrum of pathogens, graft survival, and visual prognosis. Ophthalmology 1988;95:1450-7.

71. Fontana L, Moramarco A, Mandaré E, Russello G, Iovino A. Interface infectious keratitis after anterior and posterior lamellar keratoplasty. Clinical features and treatment strategies. A review. Br J Ophthalmol 2019;103:307-4.

72. Kanavi MR, Foroutan AR, Kamel MR, Afsar N, Javadi MA. Candida interface keratitis after deep anterior lamellar keratoplasty: Clinical, microbiologic, histopathologic, and confocal microscopic reports. Cornea 2007;26:990-3.

73. Tsui E, Fogel E, Hansen K, Talbot EA, Tammer R, Fogel J, et al. Candida interface infections after descemet stripping automated endothelial keratoplasty. Cornea 2016;35:456-64.

74. Brothers KM, Shanks RMQ, Hurlbert S, Kowalski RP, Tu EY. Association between fungal contamination and eye bank-prepared endothelial keratoplasty tissue: Temperature-dependent risk factors and antifungal supplementation of optisol-gentamicin and streptomycin. JAMA Ophthalmol 2017;135:1184-90.

75. Aldave AJ, DeMatteo J, Glasser DB, Tu EY, Iliaakis B, Nordlund ML, et al. Report of the eye bank association of America medical advisory board subcommittee on fungal infection after corneal transplantation. Cornea 2013;32:149-54.

76. Hau SC, Dart JK, Vesalouma M, Parmar DN, Claerhout I, Babi X, et al. Diagnostic accuracy of microbial keratitis with in vivo scanning laser confocal microscopy. Br J Ophthalmol 2010;94:982-7.

77. Shipton C, Dard JK, Siciliano E, Luyten S, Moraillas M, Van Langen AG, et al. The clinical and microbiological profile of herpes simplex keratitis in the Netherlands. Curr Eye Res 2005;30:111-7.

78. Tu EY, Hou J. Intrastromal antifungal injection with secondary lamellar interface infusion for late-onset infectious keratitis after DSAEK. Cornea 2014;33:990-3.

79. Panda A, Pushker N, Nainiwal S, Satpathy G, Nayak N. Rhodotorula sp. infection in corneal interface following lamellar keratoplasty—a case report. Acta Ophthalmol Scand 1999;77:227-8.
80. Weng CY, Parke DW 3rd, Walter SD, Isom RF, Chang JS, Flynn HW Jr. Candida glabrata endophthalmitis transmitted from graft to host after descemet stripping automated endothelial keratoplasty. JAMA Ophthalmol 2014;132:1381-3.

81. Solomon R, Donnenfield ED, Holland EJ, Yoo SH, Daya S, Güell JL, et al. Microbial keratitis trends following refractive surgery: Results of the ASCRS infectious keratitis survey and comparisons with prior ASCRS surveys of infectious keratitis following keratorefractive procedures. J Cataract Refract Surg 2011;37:1343-50.

82. Haq Z, Farooq AV, Huang AJW. Infections after refractive surgery. Curr Opin Ophthalmol 2016;27:367-72.

83. Das S, Garg P, Mullick R, Annavajhala S. Keratitis following laser refractive surgery: Clinical spectrum, prevention and management. Indian J Ophthalmol 2020;68:2813-2818.

84. Ganesh S, Brar S, Nagesh BN. Management of infectious keratitis following uneventful small-incision lenticule extraction using a multimodal approach - A case report. Indian J Ophthalmol 2020;68:3064-6.

85. Shetty R, Kaweri L, Nujits RM, Nagaraja H, Arora V, Kumar RS. Profile of microbial keratitis after corneal collagen cross-linking. Biomed Res Int 2014;2014:340509.

86. Wilson SE, He YG, Weng J, Li Q, McDowell AW, Vital M, et al. Epithelial injury induces keratocyte apoptosis: Hypothesized role for the interleukin-1 system in the modulation of corneal tissue organization and wound healing. Exp Eye Res 1996;62:325-7.

87. Abbouda A, Abicca I, Alió JL. Infectious keratitis following corneal crosslinking: A systematic review of reported cases: Management, visual outcome, and treatment proposed. Semin Ophthalmol 2016;31:485-91.

88. Constantinou M, Daniell M, Snibson GR, Vu HT, Taylor HR. Clinical efficacy of moxifloxacin in the treatment of bacterial keratitis: A randomized clinical trial. Ophthalmology 2007;114:1622-9.

89. Gangopadhyay N, Daniell M, Weih L, Taylor HR. Fluoroquinolone and fortified antibiotics for treating bacterial corneal ulcers. Br J Ophthalmol 2000;84:378-84.

90. Khokhar S, Sindhu N, Mirdha BR. Comparison of topical 0.3% ofloxacin to fortified tobramycin-cefazolin in the therapy of bacterial keratitis. Infection 2000;28:149-52.

91. Sharma N, Arora T, Jain V, Agarwal T, Jain R, Jain V, et al. Gatifloxacin 0.3% versus fortified tobramycin-cefazolin in treating nonperforated bacterial corneal ulcers: Randomized, controlled trial. Cornea 2016;35:56-61.

92. McDonald EM, Ram FS, Patel DV, McGhee CN. Topical antibiotics for the management of bacterial keratitis: An evidence-based review of high quality randomised controlled trials. Br J Ophthalmol 2014;98:1470-7.

93. Saillard J, Spiesser-Robelet L, Gohier P, Briot T. Bacterial keratitis treated by strengthened antibiotic eye drops: An 18 months review of clinical cases and antibiotic susceptibilities. Ann Pharm Fr 2018;76:107-13.

94. Tam ALC, Cote E, Saldanha M, Lichtinger A, Slomovic AR. Bacterial keratitis in Toronto: A 16-year review of the microorganisms isolated and the resistance patterns observed. Cornea 2017;36:1528-34.

95. Ni N, Nam EM, Hammersmith KM, Nagra PK, Azari AA, Leiby BE, et al. Seasonal, geographic, and antimicrobial resistance patterns in microbial keratitis: 4-year experience in eastern Pennsylvania. Cornea 2015;34:296-302.

96. Lin A, Rhee MK, Akpek EK, Amescua G, Farid M, Garcia Ferrer FJ, et al. Bacterial Keratitis Preferred Practice Pattern®. Ophthalmology. 2019;126:P1-P55.

97. Centers for Disease Control and Prevention. Sexually transmitted diseases treatment guidelines, 2010. MMWR Morb Mortal Wky Rep 2010;59:53.

98. Acharya NR, Srinivasan M, Mascarenhas J, Ravindran M, Rajaraman R, Zegans M, et al. The steroid controversy in bacterial keratitis. Arch Ophthalmol 2009;127:1231.

99. Cohen EJ. The case against the use of steroids in the treatment of bacterial keratitis. Arch Ophthalmol 2009;127:103-4.

100. Hindman HB, Patel SB, Jus AS. Rationale for adjunctive topical corticosteroids in bacterial keratitis. Archives of ophthalmology 2009;127:97-102.

101. Srinivasan M, Mascarenhas J, Rajaraman R, Ravindran M, Lalitha P, Glidden DV, et al. Corticosteroids for bacterial keratitis: The steroids for corneal ulcers trial (SCUT). Arch Ophthalmol 2012;130:143-50.

102. O’Day DM, Head WS, Robinson RD, Clanton JA. Corneal penetration of topical amphotericin B and natamycin. Curr Eye Res 1986;5:877-82.

103. Harirprasad SM, Mieler WF, Lin TK, Sponsel WE, Graybill JR. Voriconazole in the treatment of fungal eye infections: A review of current literature. Br J Ophthalmol 2008;92:871-8.

104. Prajna NV, Krishnan T, Mascarenhas J, Rajaraman R, Prajna L, Srinivasan M, et al. The mycotic ulcer treatment trial: A randomized trial comparing natamycin vs voriconazole. JAMA Ophthalmol 2013;131:422-9.

105. Sharma S, Das S, Virdi A, Fernandes M, Sahu SK, Kumar Koday N, et al. Re-appraisal of topical 1% voriconazole and 5% natamycin in the treatment of fungal keratitis in a randomised trial. Br J Ophthalmol 2015;99:1190-5.

106. FlorCruz NV, Evans JR. Medical interventions for fungal keratitis. Cochrane Database Syst Rev 2015;CD004241. doi: 10.1002/14651858.CD004241.pub4.

107. Prajna NV, Krishnan T, Rajaraman R, Patel S, Srinivasan M, Das M, et al. Effect of oral voriconazole on fungal keratitis in the mycotic ulcer treatment trial II (MUTT II): A randomized clinical trial. JAMA Ophthalmol 2016;134:1365-72.

108. Kuriakose T, Kothari M, Paul P, Jacob P, Thomas R. Intracameral amphotericin B injection in the management of deep keratomycosis. Cornea 2002;21:653-6.

109. Sharma N, Sankaran P, Agarwal T, Arora T, Chawla B, Titiyal JS, et al. Evaluation of intracameral amphotericin B in the management of fungal keratitis: Randomized controlled trial. Ocul Immunol Inflamm 2016;24:493-7.

110. Kalarasevi G, Narayana S, Krishnan T, Sengupta S. Intrastromal voriconazole for deep recalcitrant fungal keratitis: A case series. Br J Ophthalmol 2015;99:195-8.

111. Sharma N, Agarwal P, Sinha R, Titiyal JS, Velpandian T, Vajpayee RB. Evaluation of intracameral voriconazole injection in recalcitrant deep fungal keratitis: Case series. Br J Ophthalmol 2011;95:1765-7.

112. Shelby K, Wu TG, Wilhelmus KR, Jones DB. Activity of voriconazole against corneal isolates of Scedosporium apiospermum. Cornea 2003;22:33-6.

113. Saraci MA, Erdem U, Gonlum A, Yildiran ST. Scedosporium apiospermum keratitis treated with itraconazole. Med Mycol 2003;41:111-4.

114. Edelstein S, Akduman L, Durham BH, Fothergill AW, Hsu HY. Resistant Fusarium keratitis progressing to endophthalmitis. Eye Contact Lens 2012;38:331-5.

115. Nayak N, Satpathy G, Prasad S, Vajpayee RB, Pandey RM. Correlation of proteinase production with amphotericin B resistance in fungi from mycotic keratitis. Ophthalmic Res 2010;44:113-8.

116. Tsatsos M, MacGregor C, Athanasiadis I, Moschos MM, et al. Herpes simplex virus keratitis: An update of the pathogenesis and current treatment with oral and topical antiviral agents. Clin Exp Ophthalmol 2016;44:824-37.

117. Chou TY, Hong BY. Ganciclovir ophthalmic gel 0.15% for the treatment of acute herpetic keratitis: Background, effectiveness,
tolerability, safety, and future applications. Ther Clin Manag 2014;10:665-81.
118. Wilhelmsen KR, Gee L, Hauck WW, Kurinij N, Dawson CR, Jones DB, et al. A controlled trial of topical corticosteroids for herpes simplex stromal keratitis. Ophthalmology 1994;101:1883-95.
119. Goldblum D, Bachmann C, Tappeiner C, Garweg J, Fruehe BE. Comparison of oral antiviral therapy with valacyclovir or acyclovir after penetrating keratoplasty for herpetic keratitis. Br J Ophthalmol 2008;92:1201-5.
120. Herpetic Eye Disease Study Group. Acyclovir for the prevention of recurrent herpes simplex virus eye disease. N Engl J Med 1998;339:300-6.
121. Seal D. Treatment of Sclanthamoeba keratitis. Expert Rev Anti Infect Ther 2003;1:205-8.
122. Agarwal S, Iyer G, Srinivasan B, Benurwar S, Agarwal M, Narayanan N, et al. Clinical profile, risk factors and outcome of medical, surgical and adjunct interventions in patients with *Pythium insidiosum* keratitis. Br J Ophthalmol 2019;103:296-300.
123. Agarwal S, Srinivasan B, Janakiraman N, Therese LK, S, Patel N, et al. Role of topical ethanol in the treatment of pythium insidiosum keratitis-A proof of concept. Cornea 2020;39:1102-7.
124. Das S, Sahu SK, Sharma S, Nayak SS, Kar S. Clinical trial of 0.02% polyhexamethylene biguanide versus placebo in the treatment of microsporidial keratoconjunctivitis. Am J Ophthalmol 2010;150:110-5.
125. Bryan RT, Cali A, Owen RL, Spencer HC. Microsporidia: Opportunistic pathogens in patients with AIDS. Prog Clin Parasitol 1991;1:1-26.
126. Córdova-Alcántara IM, Venegas-Cortés DL, Martínez-Rivera MA, Pérez NO, Rodríguez-Tovar AV. Biofilm characterization of Fusarium solani keratitis isolate: Increased resistance to antifungals and UV light. J Microbiol 2019;57:485-497.
127. Cordeiro RA, Portela FV, Pereira LM, Andrade AR, Sousa J, Aguiar AL, et al. Efflux pump inhibition controls growth and enhances antifungal susceptibility of Fusarium solani species complex. Future Microbiol 2020;15:9-20.
128. Zhang J, Wang L, Gao C, Zhang L, Xia H. Ocular pharmacokinetics of topically-applied ketoconazole solution containing hydroxypropyl beta-cyclodextrin to rabbits. J Ocul Pharmacol Ther 2008;24:501-6.
129. Abdul Rasool BK, Salmo HM. Development and clinical evaluation of clotrimazole–β cyclodextrin eye drops for the treatment of fungal keratitis. AAPS Pharm Sci Tech 2012;13:883-9.
130. Ustündağ-Okur N, Gökçe EH, Bozbıyık DJ, Eğrilmez S, Özer O, Ertan G. Preparation and in vitro in vivo evaluation of oloxacin loaded ophthalmic nano structured lipid carriers modified with chitosan oligosaccharide lactate for the treatment of bacterial keratitis. Eur J Pharm Sci 2014;63:204-15.
131. Wadhwa S, Singh B, Sharma G, Raza K, Katare OP. Liposomal fusidic acid as a potential delivery system: A new paradigm in the treatment of chronic plaque psoriasis. Drug Deliv 2016;23:1204-13.
132. Zhang R, He R, Qian J, Guo J, Xue K, Yuan YF. Treatment of experimental autoimmune uveoretinitis with intravitreal injection of tacrolimus (FK506) encapsulated in liposomes. Invest Ophthalmol Vis Sci 2010;51:3575-82.
133. Hu W, Metselaar J, Ben LH, Cravens PD, Singh MP, Frohman EM, et al. PEG minocycline-liposomes ameliorate CNS autoimmune disease. PLoS One 2009;4:e151.
134. Ciolino JB, Hudson SP, Mobbs AN, Hoare TR, Iwata NG, Fink GR, et al. A prototype anti fungal contact lens. Invest Ophthalmol Vis Sci 2011;52:6286-91.
135. Silva D, Sousa HC, Gil MH, Santos LF, Moutinho GM, Serro AP, et al. Antibacterial layer-by-layer coatings to control drug release from soft contact lenses material. Int J Pharm 2018;553:186-200.
136. Ito S, Nakamura J, Fukuta M, Ura T, Teshigawara T, Fukushima J, et al. Prophylactic and therapeutic vaccine against Pseudomonas aeruginosa keratitis using bacterial membrane vesicles. Vaccine 2021;39:3152-60.
137. Pineda R 2nd, Dohman CH. Adjunctive therapy and surgical considerations in the management of bacterial ulcerative keratitis. Int Ophthalmol Clin 1996;36:37-48.
138. Hamblin MR, Hasan T. Photodynamic therapy: A new antimicrobial approach to infectious disease? Photochem Photobiol Sci 2004;3:436-50.
139. Dai T, Fuchs BB, Coleman JJ, Prates RA, Astrakas C, St Denis TG, et al. Concepts and principles of photodynamic therapy as an alternative antifungal discovery platform. Front Microbiol 2012;3:120.
140. Cheron D, Verter EE, Melli S, Giseli TE, Doyle FJ Jr, Scarcelli G, et al. Collagen cross-linking using rose bengal and green light to increase corneal stiffness. Invest Ophthalmol Vis Sci 2013;54:3426-33.
141. Ting DSJ, Henein C, Said DG, Dua HS. Photoactivated chromophore for infectious keratitis-Corneal cross-linking (PACK-CXL): A systematic review and meta-analysis. Ocul Surf 2019;17:624-34.
142. Abbouda A, Abicca I, Alió JL. Current and future applications of photoactivated chromophore for keratitis-corneal collagen cross-linking (PACK-CXL): An overview of the different treatments proposed. Semin Ophthalmol 2018;33:293-9.
143. Prates R, Kato IT, Ribeiro MS, Tegos GP, Hamblin MR. Influence of multidrug efflux systems on melanin blue-mediated photodynamic inactivation of *Candida albicans*. J Antimicrob Chemother 2011;66:1525-32.
144. Atalay HT, Dogruman-Al F, Sarzhanov F, Ozmen MC, Tefon AB, Arıbas YK, et al. Effect of rifabutin/rose bengal-mediated pack-cxl on Acanthamoeba trophozoites and cysts in vitro. Curr Eye Res 2018;43:1322-5.
145. Arboleda A, Miller D, Cabot F, Taneja M, Aguilar MC, Alawa K, et al. Assessment of rose bengal versus rifabutin photodynamic therapy for inhibition of fungal keratitis isolates. Am J Ophthalmol 2014;158:64-70.e2.
146. Naranjo A, Arboleda A, Martinez JD, Durkee H, Aguilar MC, Relhan N, et al. Rose bengal photodynamic antimicrobial therapy for patients with progressive infectious keratitis: A pilot clinical study. Am J Ophthalmol 2019;208:367-96.
147. Dai T, Gupta A, Murray CK, Vrahos MS, Tegos GP, Hamblin MR. Blue light for infectious diseases: Propionibacterium acnes, Helicobacter pylori, and beyond? Drug Resist Updat 2012;15:223-36.
148. Van der Horst MA, Stalcup TP, Kaledhonkar S, Kumauchi M, Hara M, Xie A, et al. Locked chromophore analogs reveal that photoactive yellow protein regulates biofilm formation in the deep sea bacterium *Idiomarina loihiensis*. J Am Chem Soc 2009;131:17443-51.
149. Zhu H, Kocheva IE, Behlau I, Zhao J, Wang F, Wang Y, et al. Antimicrobial blue light therapy for infectious keratitis: Ex vivo and in vivo studies. Invest Ophthalmol Vis Sci 2017;58:586-93.
150. Trzaska WJ, Wigley HE, Thwaite JE, May RC. Species-specific antifungal activity of blue light. Sci Rep 2017;7:4605.
151. Former C, L’Esperance F. Argon laser phototherapy of pseudomonas corneal ulcers. Invest Ophthalmol Vis Sci 1971;10:1-8.
152. Krauss JM, Puliafito CA, Steiner RF. Laser interactions with the cornea. Surv Ophthalmol 1986;31:37-53.
153. Pellegrino F, Carrasco MA. Argon laser phototherapy in the treatment of refractory fungal keratitis. Cornea 2013;32:95-7.
154. Reitberger HH, Czugala M, Chow C, Mohr A, Burkovski A, Gruenert AK, et al. Argon cold plasma-A novel tool to treat therapy-resistant corneal infections. Am J Ophthalmol 2018;190:150-63.
155. Martines E, Brun P, Brun P, Cavazanna R, Delizian V, Leonardi A, et al. Towards a plasma treatment of corneal infections. Clin Plasma Med 2013;1:17-24.
156. Mai-Prochnow A, Clausin M, Hing J, Murphy AB. Gram positive and gram negative bacteria differ in their sensitivity to cold plasma. Sci Rep 2016;6:38610. doi: 10.1038/srep38610.

157. McDonnell G, Russell A. Antiseptics and disinfectants: Activity, action and resistance. Clin Microbiol Rev 1999;12:147-79.

158. Dagley S, Dawes EA, Morrison GA. Inhibition of growth of Aerobacter aerogenes: The mode of action of phenols, alcohols, acetone and ethyl acetate. J Bacteriol 1950;60:369-78.

159. Aqeel Y, Rodriguez R, Chatterjee A, Ingalls RR, Samuelson J. Killing of diverse eye pathogens (Acanthamoeba spp., Fusarium solani, and Chlamydia trachomatis) with alcohols. PLoS Negl Trop Dis 2017;11:e0005382.

160. Lin IH, Tseng SH, Huang FC, Huang YH. Effect of ethanol pretreatment in Acanthamoeba keratitis: A long-term follow-up study. Infect Drug Resist 2018;11:937-43.

161. Eiferman RA. Cryotherapy of Pseudomonas keratitis and scleritis. Arch Ophthalmol 1979;97:1637-9.

162. Fulhorst HW, Richards AB, Bowbyes J, Jones BR. Cryotherapy of epithelial herpes simplex keratitis. Am J Ophthalmol 1972;73:46-51.

163. Chen Y, Yang W, Gao M, Belin MW, Yu H, Yu J. Experimental study on cryotherapy for fungal corneal ulcer. BMC Ophthalmol 2015;15:29.

164. Rodriguez-Ares MT, De Rojas Silva MV, Pereiro M, Fete Sampayo B, Gallegos Chamas G, S-Salorio M. Aspergillus fumigatus scleritis. Acta Ophthalmol Scand 1995;73:467-9.

165. Meisler DM, Ludwig IH, Rutherford I, Bican FE, Langston RH, Visvesvara GS. Susceptibility of acanthamoeba to cryotherapeutic method. Arch Ophthalmol 1986;104:130-1.

166. Maumenee AE, Kornblueth W. Regeneration of the corneal stroma; review of literature and histologic study. Am J Ophthalmol 1949;32:1051-64.

167. Chi HH, Kelman CD. Effect of freezing on ocular tissues: I. Clinical and histological study of corneal endothelium. Am J Ophthalmol 1966;61:630-41.

168. Bagga B, Garg P, Joseph J, Mohamed A, Kalra P. Outcome of therapeutic deep anterior lamellar keratoplasty in advanced Acanthamoeba keratitis. Indian J Ophthalmol 2020;68:442-6.

169. Sarnicola E, Sarnicola C, Sabatino F, Tosi GM, Perri P, Sarnicola V. Early deep anterior lamellar keratoplasty (DALK) for Acanthamoeba keratitis: A randomized clinical trial. Cornea 2016;35:1-5. doi: 10.1097/ICO.0000000000000681.

170. Hill JC. Use of penetrating keratoplasty in acute bacterial keratitis. Br J Ophthalmol 1986;70:502-6.

171. Agarwal S, Iyer G, Srinivasan B, Agarwal M, Panchal S, Kumar S. Clinical profile of pythium keratitis: Perioperative measures to reduce risk of recurrence. Br J Ophthalmol 2018;102:153-7.

172. Allan BD, Dart JK. Strategies for the management of microbial keratitis. Br J Ophthalmol 1995;79:777-6.

173. Gundersen T. Conjunctival flaps in the treatment of corneal disease with reference to a new technique of application. AMA Arch Ophthalmol 1958;60:880-8.

174. Abdulhalim BE, Wagih MM, Gad AA, Boghdadi G, Nagy RR. Amniotic membrane graft to conjunctival flap in treatment of non-viral resistant infectious keratitis: A randomised clinical study. Br J Ophthalmol 2015;99:59-63.

175. Buxton JN, Fox ML. Conjunctival flaps in the treatment of refractory pseudomonas corneal abscess. Ann Ophthalmol 1986;18:315-8.

176. Chen HC, Tan HY, Hsiao CH, Huang SC, Lin KK, Ma DH. Amniotic membrane transplantation for persistent corneal ulcers and perforations in acute fungal keratitis. Cornea 2006;25:26-72.

177. Lilia A, Santander-Garcia D, Vanzzini-Zago V, Cuevas-Cancino D. Therapeutic keratoplasty for microbial keratitis: keratoplasties - surgical techniques and complications, Luigi Mosca. IntechOpen 2012;12-30. doi: 10.5772/20643.

178. Chatterjee S, Agrawal D. Recurrence of infection in corneal grafts after therapeutic penetrating keratoplasty for microbial keratitis. Cornea 2020;39:39-44.

179. Sun JP, Chen WL, Huang JY, Hou YC, Wang JJ, Hu FR. Microbial keratitis after penetrating keratoplasty. Am J Ophthalmol 2017;178:150-6.