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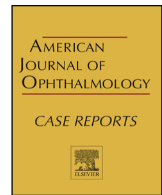

Case report

Long-term outcomes of riboflavin photodynamic antimicrobial therapy as a treatment for infectious keratitis

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A R T I C L E I N F O

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Corneal infectious keratitis
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Crosslinking
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A B S T R A C T

Purpose: To report the long-term outcomes of three patients with infectious keratitis treated with riboflavin photodynamic antimicrobial therapy (PDAT).
Observations: Case series reporting three patients with infectious keratitis unresponsive to standard medical treatment who underwent riboflavin photodynamic antimicrobial therapy (PDAT) as an adjunct therapy. One male and two female patients were treated, the median age of presentation was 58 years (range, 29–79 years). The organisms isolated and treated were *Pseudomonas aeruginosa*, *Mycobacterium chenolae*, and *Curvularia spp.* Different risk factors to develop corneal infection ulcers were identified, including corneal abrasion in a contact lens user, history of penetrating keratoplasty with chronic use of topical corticosteroids, and organic trauma. The median follow-up was 47 months (range 37–54 months), and there were no complications secondary to riboflavin PDAT treatment. Two cases underwent optical penetrating keratoplasty after infection was resolved and ocular surface was quiet for at least 3 years.
Conclusions and importance: Riboflavin PDAT can be used as an adjunct treatment in infectious keratitis to strengthen the corneal collagen fibers, delay keratolysis, and allow more time for antimicrobials to work and this way prevent a corneal perforation.

1. Introduction

Even with proper medical management, some cases of infectious keratitis can progress to a corneal perforation. In some of these cases, a therapeutic corneal transplant is required. However, performing a corneal transplant on an infected and inflamed ocular surface increases the risk of graft failure or graft rejection.[1]

Corneal crosslinking with riboflavin and ultraviolet-A (UV-A) light has been established as a first-line treatment to prevent the progression of keratoconus or corneal ectasias by strengthening the corneal collagen fibers.[2] It was later proposed as a treatment for infectious keratitis unresponsive to medical treatment and was described in the literature as photoactivated chromophore (PACK-CXL)[3,4] or riboflavin Photodynamic Antimicrobial Therapy (PDAT).[5,6] Bamdad et al. reported a randomized control trial in which the group treated with PACK-CXL had a shorter course of medical treatment and decreased need for therapeutic corneal transplantation.[7] A few articles have reported the use of riboflavin CXL as an adjunct therapy for *Pseudomonas* keratitis[4,8–10] and *Mycobacterium* keratitis,[11,12] but none have reported its utility in treating *Curvularia* spp.

We present three cases of infectious keratitis that presented to Bascom Palmer Eye Institute and were treated with riboflavin PDAT. Informed consent was obtained, and the procedure was performed following a modified Dresden protocol.[2] Under topical anesthesia,

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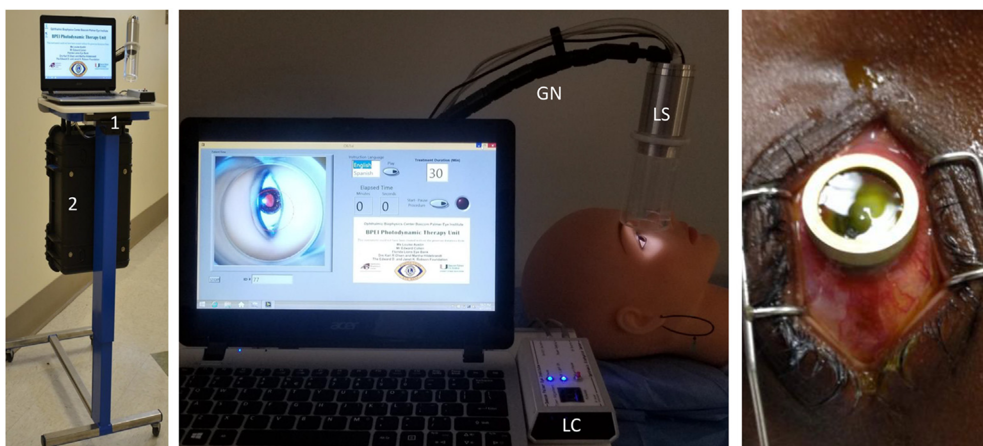

Fig. 1. Custom-built riboflavin PDAT system (A) UVA light delivery unit on stand (1: height adjustment; 2: Battery power and cooling system) (B) Laptop to adjust and monitor irradiation timing and center light source over the patient eye (LS: LED light source; LC: Light source controller; GN: goose neck clamp to adjust source over patient eye) (C) Metal corneoscleral well filled with 0.1% riboflavin.

corneal ulcer scraping was performed 1–2 mm around the corneal epithelial defect. One drop of 0.1% riboflavin in 20% dextran solution was applied to the cornea every 3 minutes for a total of 30 minutes. A custom-made shield was placed over the limbal area for protection and the cornea was irradiated for 30 minutes with a custom-made ultraviolet-A (UV-A) light source for a radiant exposure of 5.4 J/cm^2. (Fig. 1). The light source, previously described by Halili et al., who conducted *in vitro* experiments with multiple organisms, contains twenty-four 375nm LEDs and emits a power density of 3 mW/cm^2.[6]

2. Findings

2.1. Case 1

A 29-year-old female was referred to our institution with a corneal ulcer secondary to a corneal abrasion while removing a soft contact lens from her right eye. Corneal ulcer cultures came back positive for *Pseudomonas aeruginosa* with S-/U+ genotype which did not respond to standard medical treatment for 15 days (Table 1 and Fig. 2A). The patient underwent riboflavin PDAT with placement of an amniotic membrane (*Ambiodisk, Costa Mesa, CA, USA*) and bandage contact lens following the procedure. Topical moxifloxacin with Doxycycline and Vitamin C was continued, while the patient started tapering prednisolone acetate.

The patient significantly improved after 2 weeks of riboflavin PDAT (Fig. 2 B). Two months after PDAT, antibiotic treatment was suspended. One year after PDAT, the best corrected visual acuity (BCVA) was 20/800, and examination revealed a vascularized and opacified cornea with complete epithelialization and quiet conjunctiva without hyperemia (Fig. 2C). Fifteen months after riboflavin PDAT treatment, we proceeded with an optical penetrating keratoplasty (PK) followed by amniotic membrane placement without complications (Fig. 2D and E). Four years and 6 months after the riboflavin PDAT procedure, BCVA was 20/30, IOP was 20 mmHg, and the corneal graft remained clear without signs of rejection or infection. (Fig. 2F).

2.2. Case 2

A 79-year-old female patient with history of PK secondary to Fuchs corneal dystrophy 13 years prior, presented with left eye progressive infectious keratitis in the left eye presumed to be secondary to a loose suture. Corneal ulcer cultures came back positive for *Mycobacterium chenolae* sensitive to Clarithromycin and with intermediate sensitivity to Amikacin (Table 1 and Fig. 3A). There was no improvement in the following 7 days on standard medical treatment and the decision to undergo riboflavin PDAT was made due to the presence of progressive keratolysis. After PDAT, she was started on 0.5% Cyclosporine drops 4 times a day. Nine days after PDAT, the BCVA was 20/200, there was decreased conjunctival hyperemia, and the epithelial defect had healed (Fig. 3B). Three years after riboflavin PDAT, the patient continued on acetate prednisolone eye drops once a day, her BCVA was 20/40 and IOP was19 mmHg. Slit-lamp examination showed a quiet conjunctiva, clear corneal graft and IOL in place (Fig. 3C).

2.3. Case 3

A 58-year-old male presented with a corneal ulcer in the right eye caused by injury while cutting a tree branch. Corneal ulcer cultures came back positive for *Curvularia* spp. The patient was started on Natamycin every hour and 0.5% Moxifloxacin every 4 hours (Table 1 and Fig. 4 A). After 15 days without clinical improvement and due to rapid progression of the stromal necrosis, the patient underwent riboflavin PDAT. After treatment, the medication regimen was: Natamycin eye drops every 2 hours, 200 mg Fluconazole tablets twice a day, Cyclosporine-A eye drops 4 times a day, Atropine once a day, and Moxifloxacin (Fig. 4B). After 2 months, the corneal infiltrate disappeared,

Table 1
Summary of patients with infectious keratitis who underwent riboflavin photodynamic antimicrobial therapy. All patients underwent a single PDAT session.

Case #	Organism	Treatment (Time on Tx prior RB PDAT)	Time to Clinical Resolution (weeks)	Complication	Surgical Treatment (time post PDAT)	Time FU (months) Comment
1	*Pseudomonas aeruginosa*	0.5% Moxifloxacin 14 mg/ml Fortified tobramycin Doxycycline PO 100 mg BID **(15 days)**	8	None	PK (12 m)	54 Corneal graft clear
2	*Mycobacterium chenolae*	5% Amikacin 2% Clarithromycin 0.5% Moxifloxacin **(7 days)**	4	None	None	36 Cornea clear
3	*Curvularia spp.*	5% Natamycin 200 mg Fluconazole tablets, 0.5% Moxifloxacin **(15 days)**	10	None	PK (31 m)	41 Corneal graft clear

PK: penetrating keratoplasty; RB PDAT: riboflavin photodynamic antimicrobial therapy; FU: follow-up.

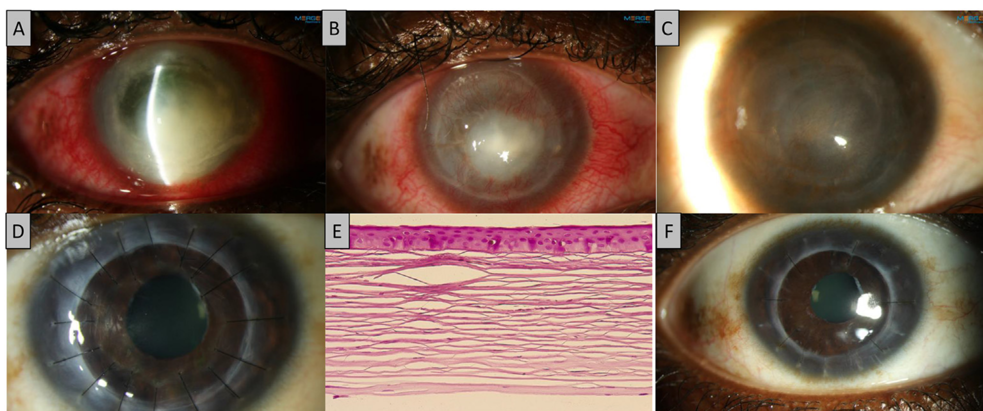

Fig. 2. Case 1. (A) Slit-lamp photograph of the right eye with corneal melting inferiorly and thinning. (B) two weeks after riboflavin PDAT, presenting with central corneal infiltrate shrinkage more than 50% and increased peripheral corneal neovascularization. (C) One year after riboflavin PDAT, no corneal infiltration or diffuse corneal scarring were observed. (D) Optical penetrating keratoplasty (OPK) was done one year after PDAT. (E) No organisms identified on gram stained section of cornea. Brown and Hopps gram stain, 400X. (F) OPK remains clear on last follow-up. . (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

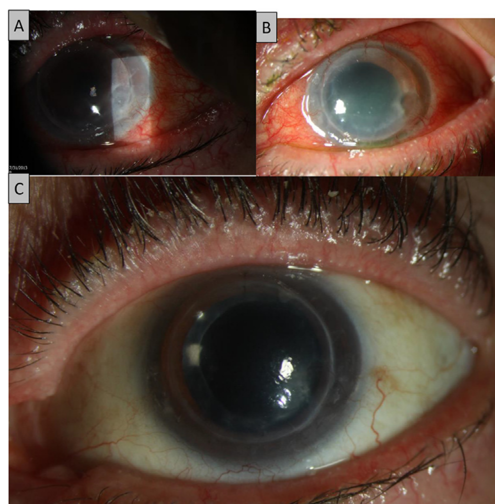

Fig. 3. Case 2. (A) Slit-lamp photograph of the left eye. Corneal infiltrate of 1 mm at 3–4 o'clock with corneal epithelial defect. (B) Nine days after riboflavin PDAT, corneal melting stopped, and epithelium healed. (C) Four weeks after riboflavin PDAT, no evidence of infiltrate, corneal scar at 3–4 o'clock. (D) Corneal graft remains clear on last follow-up.

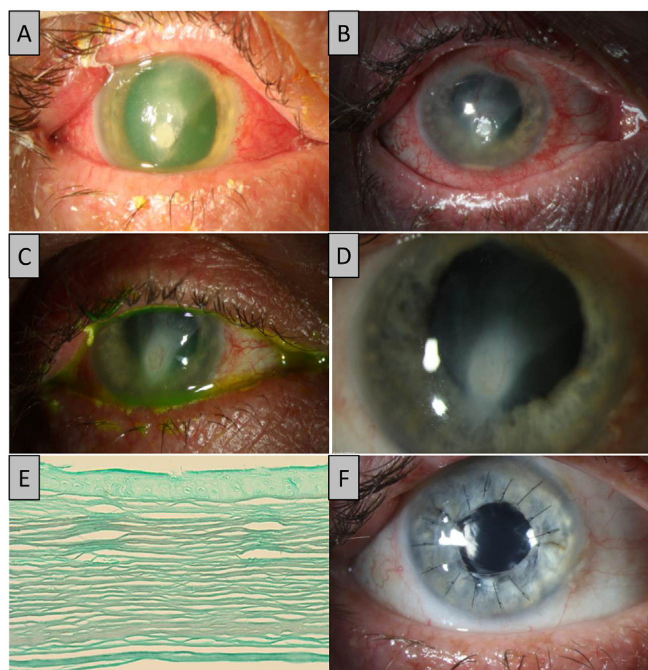

Fig. 4. Case 3. (A) Slit-lamp photograph of the right eye. Initial presentation: central corneal epithelial defect and dense inferior paracentral corneal infiltrate. (B) Four weeks after riboflavin PDAT, corneal infiltrate decreased and began developing central neovascularization with 1 mm hypopyon. (C) After 2 months, healed cornea with no infiltrate or corneal epithelial defect. (D) Six months after riboflavin PDAT, right eye revealed corneal scarring.(E) No organism identified on Gomori Methenamine-Silver Nitrate stain (GMS) stained section of cornea 400 X. (F) OPK remains clear on last follow-up.

but evidence of deep stromal neovascularization was found (Fig. 4C). Topical and systemic antifungal medications were discontinued. Six months after riboflavin PDAT, the patient's visual acuity was 20/800 with moderate corneal scarring in the visual axis, a cataract, and iris synechiae (Fig. 4D). Finally, the patient underwent cataract surgery and intraocular lens placement without complications. Four months after surgery, BCVA was 20/70, but the patient complained of severe glare. An optical PK and pupilloplasty was performed (Fig. 4 E, and F). Three years and 5 months after riboflavin PDAT, BCVA was 20/70, IOP was 16 mmHg, and the corneal graft remained clear with no signs of rejection or infection.

3. Discussion

Infectious keratitis is a challenging disease, where the causative organisms may display unpredictable behavior and resistance to standard medical treatment. Furthermore, complications that arise from the resistance to topical antimicrobials can lead to devastating consequences.[4] The patients in this study exhibited risk factors of infectious keratitis: contact lens use, prior corneal surgery, and prolonged steroid use.[13],[14],[15]

Over the last decade, attempts have been made to treat refractory cases of infectious keratitis with new medications and technologies, one such being PACK-CXL. Both *in vitro* and *in vivo* studies have shown microbial inhibition of *Pseudomonas aeruginosa*,[16–18] *Mycobacterium chenolae*,[12] and filamentous fungi with CXL treatment.[19]

Curvularia spp. is a rare cause of fungal keratitis in the United States, however, there has been an increase of in the number of cases reported in our institution.[20] In the case reported, the patient progressed to stromal necrosis despite compliance to standard medical treatment. Other cases have either responded to standard medical treatment, required a therapeutic penetrating keratoplasty, or have been treated for endophthalmitis.

Studies have shown that the adjunct effect of antibiotics and CXL is greater in treating infections than antibiotics alone or CXL alone.[17] The patients in our study may be benefitting from this finding, and the marked improvement after PDAT may be due to the synergistic effect of the light therapy and medications. Given the aggressive nature of the microorganisms and the advanced stage of the infections, adjunct treatment was offered to the patients to provide the most effective

treatment.

Studies by Said et al. and Kasetsuwan et al., showed that PACK-CXL reduced late complications such as corneal perforation or recurrence of infection compared to antibiotic treatment alone.[4,21] Moreover, Bamdad et al. demonstrated a faster recovery of epithelial defect and infiltrates with PACK-CXL.[7] Zamani et al. reported 8 patients diagnosed with *Pseudomonas aeruginosa* keratitis who did not respond to standard antimicrobial treatment; however, after riboflavin CXL treatment, all patients had substantial improvement.[22] This is similar to our cases, in which patients continued to worsen despite standard medical treatment until riboflavin PDAT was performed. Shortly after PDAT, patients improved both subjectively and objectively, reporting less pain and appearing better on slit lamp exam. Our presented cases have a significant longer follow up after riboflavin PDAT compared to reported cases in the literature.

All three patients treated with riboflavin PDAT had a good long-term outcome with no complications. A potential explanation for this may be increased resistance of corneal tissue to enzymatic digestion following PDAT. This enhances corneal strength, delays corneal melting, and allows time for the antimicrobials to take effect.[23] Further studies should be performed to better understand the changes of tissues following PDAT.

In summary, the cases presented highlight the use of riboflavin PDAT as an adjunct treatment for infectious keratitis, with good long-term outcome. Even in the setting of thin corneas, this treatment can help prevent a perforation that would normally result in a therapeutic corneal transplant. Although results are encouraging, and the patients presented had favorable outcomes, we understand the limitations of a retrospective study. Prospective controlled clinical trials are needed to confirm the effectiveness of Riboflavin PDAT in severe cases of infectious keratitis.

4. Patient consent

The project was deemed to meet criteria for a case series by the University of Miami Institutional Review Boards (IRB). Therefore, no IRB submission was required prior to reviewing the cases. The study was conducted in accordance with the principles of the Declaration of Helsinki. Patients in this series provided signed voluntary and informed consent to the described treatment. Patients in this series displayed appropriate capacity to provide consent. Patients understood the risks, benefits, and alternatives for the riboflavin photodynamic antimicrobial therapy and understood they were entitled to withdraw previous consent at any time during the treatment.

Consent to publish the case series was not obtained from the patients. The case series does not contain any identifying information.

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Conflicts of interest

None of the authors have financial disclosures.

Authorship

All authors attest that they meet the current ICMJE criteria for Authorship.

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