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Original Research Article

Uncommon Microorganisms in Corneal Ulcers: Case Series from a Tertiary Care Centre in Central Kerala.

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Abstract

Corneal ulcer is an epithelial defect with infiltration of underlying stroma and tissue necrosis. It is a potentially sight threatening infection caused by bacteria, fungus, virus, and parasites. Fungi commonly responsible are Aspergillus, Candida and Fusarium. Common bacteria causing ulcer include Staphylococcus, Pseudomonas, Pneumococcus, Enterobacteriacea and Neisseria. Corneal ulcer may be caused by other not so common microorganisms whose detailed case description is seen only rarely. Rare bacterial pathogens that cause corneal ulcer include atypical Mycobacteria, Nocardia spp., Chryseobacterium spp., Enterobacter spp., Bartonella henslae. Rare fungi causing corneal ulcer include Alternaria spp., Acremonium spp., Cladosporium spp., Curvularia spp., Pseudallescheria spp., and others. Pathogens vary among geographical locations depending on the local climate. The clinical presentation, microbiological picture and response to empirical therapy may be different for corneal ulcer with these uncommon organisms. We describe a case series of patients with corneal ulcer caused by not so common organisms in a period of four and a half years from July 2018 to January 2023 that presented to a tertiary care teaching hospital at central Kerala. The data was retrieved from the corneal ulcer register maintained at the cornea clinic of Department of Ophthalmology at the study institute. Photos were retrieved from stored data of the computer and were anonymized. Details including history, clinical examination findings, investigation results, treatment given and follow up details entered in the register was used for filling proforma. Some cases may progress rapidly in the absence of specific therapy leading to perforation and visual loss, which can be prevented by early identification of causes and specific therapy. Knowledge on the uncommon causes of corneal ulcer describing its clinical features and differences from the more common causes of corneal ulcer will help in managing future similar cases.

Introduction

Corneal Ulcer (CU) is the most common cause of monocular corneal blindness worldwide. [1] Ocular trauma and CU cause an estimated 1.5 to 2 million new cases of corneal blindness annually. [2] Nearly 90% of the global cases of ocular trauma and corneal ulcers that lead to corneal blindness occur in developing countries [3] There are an estimated 1.22 lakh bilateral and 10.98 lakh unilateral corneal blind in India and an estimated 25,000 to 30,000 cases of corneal blindness occur every year in India. [4,5] The estimated prevalence of corneal blindness in the Indian population is 0.45% (95% CI: 0.27-0.64%). [6] Gonzalez, et al reported that the annual incidence of CU in Madurai District of South India was 1,130 per million population. [6]

CU is a potentially sight threatening infection caused by bacteria, fungus, virus and parasites. Several studies from India have reported fungal (59.09%) and bacterial CU (19.31%) as the most common types of CU with regional variations in the aetiological prevalence of CU. [7] Trends in bacterial and fungal keratitis in south India from 2002–2012 have shown 34.3% fungus and 24.7%

bacteria as aetiological agents. The most common bacterial cause is Streptococcus pneumoniae (7%) followed by Pseudomonas aeruginosa (5.4%), Nocardia spp. (1.6%), and Staphylococcus aureus (1.2%). The most common fungal causes are Fusarium spp. (14.5%) and Aspergillus spp. (8.8%) of the cultures. [8] Another study from south India showed 47.1% bacterial and 46.8% fungal culture positivity. The most common bacterial pathogen isolated are Streptococcus pneumoniae 44.3% and Pseudomonas spp. (14.4%). The most common fungal isolates are Fusarium spp. 47.1% and Aspergillus spp. (16.1%). [9] Another study reported microbial aetiology as bacterial in 44.5% and fungal in 49.5% cases. The most common fungus isolated was Fusarium (31%) followed by Aspergillus spp. (11%). Common bacterial isolates are Staphylococcus aureus (18%) followed by Streptococcus pneumonia, Pseudomonas aeruginosa and Klebsiella pneumonia. [10]

A study from central Kerala reported bacterial isolates in 52.08% and fungal isolates in 47.92%. Bacterial isolates include Staphylococcus epidermidis (32%), Streptococcus pneumoniae (16%), Pseudomonas aeruginosa (16%), Staphylococcus aureus (12%). Fungal isolates were mainly Fusarium spp. 39.13%, Aspergillus spp. And Curvularia spp. (13.04% each) [11] Another study from Kerala showed equal frequency of bacterial and fungal infections (36% each). Predominant fungal pathogens included Aspergillus spp. (24%) followed by Candida albicans (8%) and the predominant bacterial pathogens were Streptococcus pneumoniae (16%) and Pseudomonas (8%). [12] Another study from south Kerala reported 27.41% bacterial and 69.78% fungal cases. Among bacteria, Pneumococci and Pseudomonas were predominant (26.14% each) and Fusarium (37.05%) was the most frequent among Fungi. [13] Rare bacterial pathogens that cause CU include Atypical mycobacteria, Nocardia spp., Chrysebacterium spp., Enterobacter spp., Bartonella henslae and rare fungi causing CU include Alternaria spp., Acremonium spp., Cladosporium spp., Curvularia spp., Pseudallescheria spp., and others. [14]

Pathogens vary among geographical locations depending on the local climate. The clinical presentation, microbiological picture and response to empirical therapy may be different for CU with these uncommon organisms. Some cases may progress rapidly in the absence of specific therapy leading to perforation and visual loss, which can be prevented by early identification of causes and specific therapy. However, these uncommon organisms are difficult to identify by routine culture methods. A high index of suspicion along with proper history and thorough knowledge about these organisms is needed to identify these organisms. Special investigations like a polymerase chain reaction, enzyme-linked immunosorbent assay, gene sequencing, and mass spectroscopy may be necessary. [(14)] Reports on uncommon organisms causing CU are limited. A case report on fungal keratitis due to Cylindrocarpon lichenicola was reported from Kasaragod, Kerala [15] and a case report on Nocardia puris keratitis was previously reported from Kerala. [16] We report a series of not so common or uncommon causes of CU from central Kerala describing its clinical features and differences from the more common causes of CU.

Materials and Methods

We describe a case series of patients with corneal ulcer caused by not so common or rare causative organisms in a period of two years from October 2018 to September 2020 that presented to a public sector tertiary care teaching hospital at central Kerala. The data was retrieved from the corneal ulcer register maintained at the cornea clinic of the Department of Ophthalmology at the study institute. Photos were retrieved from stored data of our computer and were anonymized. Details including occupation, presenting complaints, history of trauma, systemic comorbidities and topical steroid use were retrieved from the register. All patients with corneal ulcer had undergone a detailed ocular examination using a slit lamp biomicroscope, Best Corrected Visual Acuity (BCVA) and syringing of the duct. The edge and base of the ulcer were scraped using a sterile number 15 blade. The contents were transferred to two glass slides for gram stain and a 10% Potassium Hydroxide (KOH) wet mount for direct microscopy and inoculated to Blood Agar (BA) for culture and sensitivity in a C shaped fashion and evaluated at 24 and 48 hours. Inoculation on Sabouraud Dextrose Agar (SDA) was done for fungal culture that were examined daily for up to two weeks.

Bacterial pathogens were specifically identified using the microscopic morphology, staining characteristics and biochemical properties. Species identification was also done using VITEK automated machine. Fungal filaments were detected by microscopy of KOH wet mount. Fungi were identified by the colony characteristics on SDA and by morphological appearance of spores on Lactophenol cotton blue (LPCB) stain. All laboratory methods were done following standard protocols. Microbial cultures were considered positive only if any of the following criterions were met (a). The growth of the same organism was demonstrated on two or

more solid media on the C- streak; or semi confluent growth at the site of inoculation on one solid medium, (b) Is consistent with clinical signs, and (c) smear results were consistent with cultures. Empirical therapy was started with fortified Vancomycin 50 mg/ml (5%) and Amikacin 40 mg/ml eyedrops hourly for the 1st two days, then two hourly for next five days and then tapered according to clinical symptoms. Homatropine 2% eyedrops was given thrice daily and supplementation of Vitamin C 500 mg orally once daily was started in all cases. Once fungal filaments were seen in 10% KOH mount, Natamycin 5% eye drops was started one hourly for first two days, then 2 hourly for next one week and then tapered according to clinical response and continued for up to two months.Topical antiglaucoma medications was prescribed in all cases with hypopyon. Any changes made in treatment will be specifically mentioned while describing individual cases. Specific medical therapy based on culture and sensitivity report and necessary surgical treatments (punctum cautery, AMT, Tarsorrhaphy, PKP) were given based on the clinical condition and course of the CU. Follow up was done till resolution of the ulcer or corneal opacity was seen. The CU was defined as resolved if epithelial defect was healed with non-progression of corneal infiltrate and eye is quiet with no signs of inflammation. The reason and stage of the dropout from the study was documented.

Case Reports

Case 1

Case 1 was a 63-year-old unemployed poorly built and nourished one eyed female who was on radiotherapy and chemotherapy for carcinoma maxilla for the past two years. Second cycle of chemotherapy was stopped in between because of poor health. The patient gave a history of urinary tract infection and a history of eye rubbing and blurring of vision. The attender had noticed a white discoloration of her right eye (RE) with profuse discharge and redness two weeks back. She was treated with topical antibiotic drops (reconstituted Cefotaxime) at a local hospital and was referred to the study institute as the ulcer was not responding. There was no history of Diabetes Mellitus. On examination, the best corrected visual acuity (BCVA) was 1 meter counting finger (mCF) RE with profuse greenish discharge. Conjunctival congestion (CC)and circumcorneal congestion (CCC) was present with a 6 x 5 millimetre(mm) greenish ulcer with infiltrate and abscess up to mid stroma with 0.5 mm hypopyon. Patient was started on Fortified Vancomycin and Amikacin eye drops empirically along with Homatropine eyedrops in the dosage as described earlier. Timolol 0.5 % eyedrops was prescribed twice daily. There was regurgitation of clear fluid through lower punctum and punctum cautery was done. Gram stain revealed gram negative coccobacilli (GNCB) and culture report from automated VITEK machine showed Oligella ureolyticum sensitive to Cefotaxime, Ciprofloxacin and Gentamicin. Specific therapy based on the culture reports was given with systemic Ciprofloxacin 500 mg twice daily for one week along with topical fortified Gentamicin 14 mg/ml (1.4%) eyedrops one hourly initially for the first two days, then two hourly for five days and then tapered depending on clinical response. As she looked highly malnourished, she was given nutritional supplements both in form of food items like milk and egg and tablets like Vitamin C, B complex and capsule A and D each in a once daily dosage. Discharged after two weeks as the ulcer started responding. Review at one month showed 3x3 epithelial defect with decreasing stromal abscess and no hypopyon. Further review after two months shows central macular corneal opacity. Final BCVA was 3mCF as she had age related cataract in that eye.

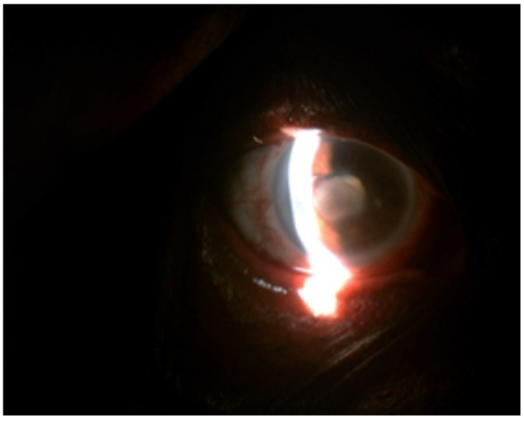

Figure 1: Oligella ureolytica CU Slit lamp picture at the healing stage.

Case 2

A 78-year-old unemployed male presenting with redness, watering, pain left eye (LE) of one week duration and white discoloration for three days. The patient had a history of loss of vision LE two years back following injury with thorn and a history of bronchial asthma for 30 years with inhaler treatment. The patient had consulted a local hospital and was started on antibiotics and antifungal eyedrops. On examination, BCVA was no light perception (PL). LE showed CC and CCC and an epithelial defect of 5x 4 mm with stromal infiltrate up to mid stroma with inferior corneal thinning and surrounding corneal oedema with DM folds. A Three mm hypopyon was present. The lacrimal duct was free. Empirical therapy with fortified antibiotics was started as already mentioned. Tab Acetazolamide 250 mg twice daily was started along with Dorzolamide 20 mg/ml and homatropine 2% eyedrops thrice daily. On microscopy, fungal filaments were seen in 10 % KOH and Penicillium species was identified in culture. Natamycin 5% eye drops was started one hourly for first two days, then two hourly for next two weeks and then tapered according to clinical response and continued for up to two months. Amniotic membrane grafting (AMT) was done after 1 week of starting Natamycin eye drops as the ulcer was not healing. Ulcer started healing after AMT. Review at 1 month showed signs of healing like vascularization and healing of epithelial defect and reduced size of infiltrate. At two months, a corneal opacity had formed. Final visual acuity was PL negative as before.

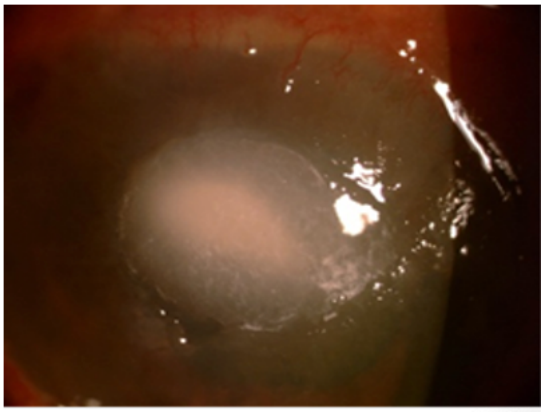

Figure 2: Penicillium CU slit lamp picture in the healing stage with vascularisation on to cornea.

Case 3

A 57-year-old male labourer presented with watering, pain, blurring of vision, redness and discharge RE of three days duration following falling of mud in the eye. Patient was on topical antibiotics from local hospital. On examination, vision in the RE was 6/ 60, the duct was free and lid oedema and discharge was present. CC, CCC with 2x1 mm epithelial defect with anterior stromal infiltrate with feathery margins and surrounding corneal oedema with DM fold was present. The patient was treated as an outpatient case with Natamycin 5% eyedrops one hourly for first two days, then two hourly for next two weeks as fungal filaments were seen in KOH wet mount. Moxifloxacin 0.5% eyedrops, Homatropine eyedrops and Tab vitamin C also was started. The patient was symptomatically better in the next two visits done after three and six days. CC and CCC had decreased but epithelial defect and infiltrate remained the same. The surrounding cornea was clear. Culture report showed Chrysosporium species with septate filamentous spore on Lactophenol cotton blue (LPCB). After one week, the patient presented with 2 mCF vision and 4x3 mm epithelial defect with stromal abscess, ring infiltrate, DM folds and 1.5 mm hypopyon. He was admitted and started on systemic Tab Voriconazole 200 mg twice daily for 14 days and topical Voriconazole 1% eyedrops two hourly for two weeks and tapered according to clinical signs. Intra stromal injection of Voriconazole 50 microgram/0.1 ml given twice, on day two and four of admission. Moxifloxacin 0.5% eyedrops and Atropine 1% eye ointment was advised thrice daily. Timolol eyedrops was prescribed twice daily along with Vitamin B and C tablets each once daily. Subconjunctival Mydricaine injection was given thrice during two weeks hospital stay. Discharged after two weeks as the ulcer started healing. In next 2 visits 1-week apart epithelial defect started decreasing in size with static hypopyon. At 1-month stromal infiltrate started decreasing in size along with epithelial defect. At 2 months a pigmented macular opacity had formed. Final BCVA was 6/36.

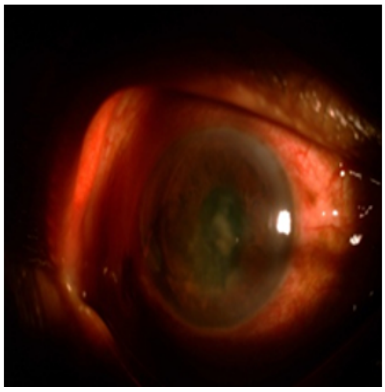
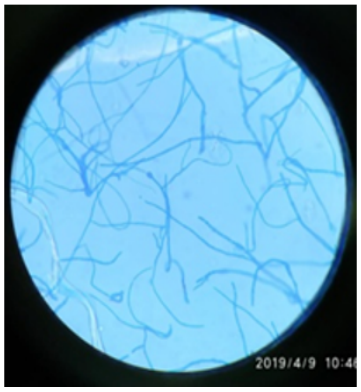

Figure 3 a **Figure 3 b** **Figure 3 c**

Figure 3: (a) Chrysosporium CU slit lamp picture. (b) LPCB showing septate filamentous spore. (c) Culture of Chrysosporium species.

Case 4

This was a 70-year-old unemployed male with complaints of pain, redness, watering LE of four days duration following finger nail injury. The patient was started on systemic and topical antibiotics after consultation at a local hospital. The patient was a known case of pseudo exfoliation glaucoma LE for past five years on Bimatoprost and Timolol eye drops and a history of ocular surgery post trauma LE and history of hypertension. On examination, the vision was PL positive in LE and discharge was present. Duct showed full regurgitation of clear fluid through upper punctum. CC, CCC with epithelial defect of 10 x 4 mm and stromal abscess of 8x4 mm with inferior corneal thinning and hypopyon of 0.5 mm was seen. Patient was started on empirical F. Vancomycin and Amikacin e/d, Atropine e/o, Dorzolamide e/d and Vitamin C in the dosages already mentioned and Tab. Ciprofloxacin 500 mg twice daily for 5 days. Gram stain showed two to three pus cells and gram-positive filaments. Acid fast bacilli was seen in Modified Ziehl Neelsen technique. Culture showed Nocardia species sensitive to Septran and Amikacin. The species identified as Nocardia Puris from a higher centre with gene sequencing technology. After starting specific treatment with Tab. Septran DS twice daily, the CU started healing and patient was discharged after 10 days. During the next review, after 1 week, CU showed signs of healing like superficial vascularization on to cornea and the patient was better symptomatically. But the epithelial defect was persisting with BCVA of 2/60. Hence AMT and Tarsorrhaphy done to promote healing of epithelial defect. But on next visit after one week, patient complained of worsening of symptoms with increased pain, watering, discharge, redness for past 3 days. On enquiry he had stopped taking Tab. Septran three days back. The patient was readmitted, but the ulcer worsened to perforation, total Iris prolapse and pseudo cornea formation even after restarting Tab. Septran and I/V Amikacin. An evisceration was finally done.

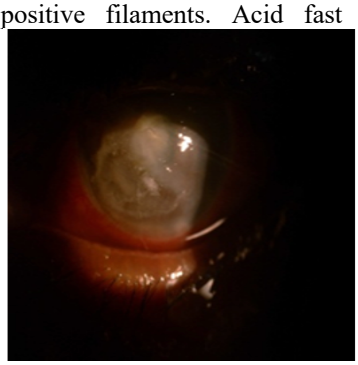
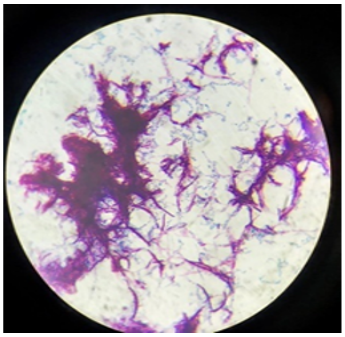
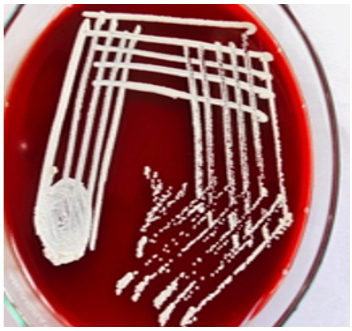

Figure 4a **Figure 4b** **Figure 4c**

Figure 4: (a) Nocardia puris CU slit lamp picture. (b) Ziehl Neelsen staining showing acid fast bacillus. (c) Culture showing growth of Nocardia puris.

Case 5

A 48-year-old male labourer presented with complaints of pain, redness, foreign body sensation, photophobia and blurred vision of 3 days duration following exposure to wooden dust particles in the LE. The patient had a history of LMN facial palsy (left) since childhood. He gave a history of starting topical antibiotics from the local Hospital and a history of topical steroid use for fall of cement particle LE. On

examination, vision LE was 6/12 and the duct was free. Lagophthalmos was present with minimal lid oedema. Initial presentation resembled viral keratitis with CC, CCC, branching hyphae appearance, keratic precipitates (KPs) and corneal oedema. Patient was empirically started on topical Acyclovir 3% eye ointment 5 times daily along with Artificial tear suppliments two hourly. But in the next visit after one week, CU was seen worsened with epithelial defect of 4 x 3 mm with stromal infiltrate up to posterior stroma with feathery margins, surrounding corneal oedema, endothelial plaque, KPs, 2 mm hypopyon and the BCVA was reduced to 5/ 60, and the patient was admitted. Acyclovir was stopped and patient was started on fortified Vancomycin and Amikacin eye drops, Timolol e/d, Atropine e/o and Vit C. Flurbiprofen eyedrops was prescribed four times daily. Microscopy showed fungal filaments in 10 % KOH. Culture showed Phaeoacremonium species. The patient was started on Natamycin 5% eye drops one hourly for first two days, then 2 hourly for next one week and then tapered according to clinical response and continued for up to two months. After 1-month, the corneal ulcer was healing with no hypopyon. By 2 months, a macular corneal opacity with thinning temporally and pigmented KPs was seen. Final BCVA was 6/12 in LE.

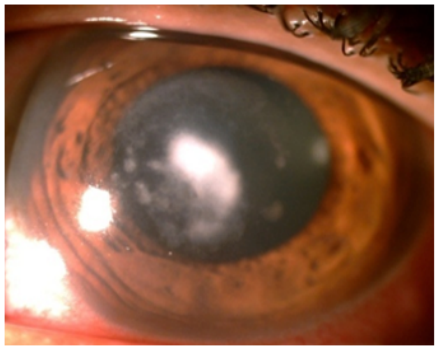

Figure 5: Phaeoacremonium CU initial presentation slit lamp picture.

Case 6

A 48-year-old male labourer presented with pain, redness, watering, defective vision LE for the past four days following fall of cement particle one week back. On examination, BCVA was hand movements in the LE and lid oedema and discharge was present. CC, CCC, and a 5x5 mm epithelial defect with stromal infiltrate up to mid stroma with central thinning, surrounding corneal oedema with DM folds, 3 mm hypopyon and membrane in anterior chamber was seen. The duct was free. Patient was started on empirical fortified Vancomycin and Amikacin eyedrops, Timolol e/d, Atropine e/o in the dosages already mentioned and Tab. Acetazolamide 250 mg twice daily. KOH showed fungal hyphae. Culture showed Cylindrocarpon lichenicola. Voriconazole e/d and Natamycin eyedrops each two hourly daily was prescribed and was advised penetrating keratoplasty. But patient went against medical advice and was lost to follow up.

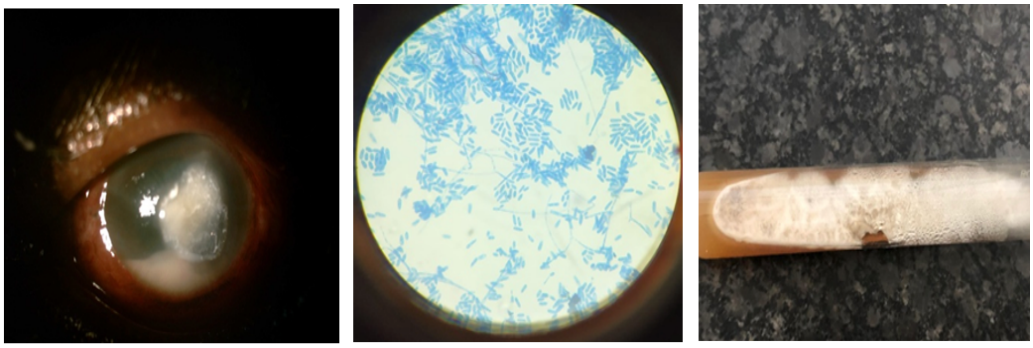

Figure 6a **Figure 6 b** **Figure 6c**

Figure 6: (a) Cylindrocarpon lichenicola CU slit lamp picture. (b) LPCB microscopy picture. (c) Culture showing growth.

Case 7

A 51-year-old male farmer presented with history of thorn prick RE 6 days back and complaints of defective vision, redness, pain, white discoloration. The patient gave a history of putting a proprietary ayurvedic topical eye preparation - Ilaneer kuzhambu. On examination, the vision in RE was 2mCF. CC, CCC, 6x3mm

epithelial defect with 3x3 mm stromal abscess, corneal oedema, DM folds, exudative membrane in anterior chamber and 1 mm hypopyon was present. Started empirically on F. Vancomycin and Amikacin e/d, timolol e/d and homatropine e/d. Microscopy showed fungal filaments and culture showed Curvularia species. Started on Natamycin 5% eyedrops was started one hourly for first two days, then 2 hourly for next one week and then tapered according to clinical response and continued for up to two months. Discharged after 4 days as patient symptomatically better and vision improved to 6/36 with 2x2 mm epithelial defect and 4x3 mm stromal infiltrate and no hypopyon. Review after 1 week showed BCVA 6/18. Epithelial defect and stromal infiltrate were status quo. After 2 weeks, the ulcer started healing with 2 x 2 mm epithelial defect and a 3x3 mm stromal infiltrate up to anterior stroma with no hypopyon. Review after 1 month showed healing CU with decreasing anterior stromal infiltrate. Review at two month showed nebular corneal opacity with BCVA of 6/12.

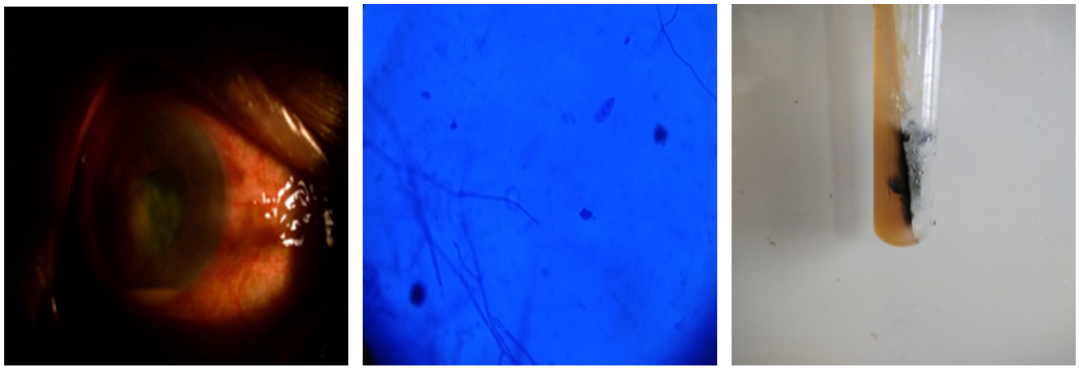

Figure 7a **Figure 7b** **Figure 7c**

Figure 7: (a) Curvularia CU slit lamp picture. (b) LPCBshowing typical conidia. (c) Culture showing typical black pigment.

Discussion

Fungal keratitis is more common than bacterial keratitis in Tropical countries and is a diagnostic and therapeutic challenge. Delayed clinical diagnosis is because of lack of suspicion and the slow growth of fungus which increases the time for confirming diagnosis. Contamination can occur as they are present in environment or body as commensals and so their role as pathogens is difficult to ascertain. Predisposing factors include trauma, topical steroid use, traditional eye medicine use. Incidence of fungal keratitis is 34.4% in India. 7.3% in north India, 32% in east India, 38.9 % in west India and 32-39.8 % in south India. Fusarium is most common in south India while Aspergillus is more common in rest of India. Dematiaceous fungi are common in tropical regions. [17]

This is the first reported case of a CU caused by Oligella ureolytica. Oligella ureolytica causing conjunctivitis is reported in literature. [18] It is a motile aerobic gram-negative coccobacillus (GNCB) found as a commensal organism in human urinary tract and is difficult to isolate by conventional lab procedure. Five cases of pathogenic infection with Oligella ureolyticum described in literature were in patients that ranged from new-borns to 89-year-old patients from India, Turkey, Canada and US. All cases resolved with drug treatment. Of the reported cases, all occurred as opportunistic infections in patients with a source of immunosuppression such as malignancy, HIV or new born status, also malnutrition, tobacco use and advanced age. [19] Our case was a malnourished carcinoma maxilla patient with history of tobacco chewing and UTI and the organism may have spread through contaminated urine to eyes by the frequent eye rubbing and minor trauma caused by it. We were suspecting Pseudomonas by the greenish discoloration of ulcer and discharge. We identified it as Oligella ureolytica by GNCB on gram smear from corneal ulcer and species identification by VITEK. The colonies may appear as small white opaque with continuous margins in culture. The ulcer started healing once we supplemented nutrition along with antibiotics. A limitation is that identification of this bacterium is difficult with commonly available lab procedures and the incubation period is long (4 days). We believe many cases of Oligella ureolyticum infection may have gone unrecognized or were incorrectly identified. Some may have been dismissed as contamination because of lack of familiarity with these bacteria. [19] Problem appears to be more profound when microbial identification systems are used to describe new infectious syndromes with previously unrecognized pathogens. [20] Our review suggests that advancements in laboratory techniques will lead to more recognition of cases

and that further studies may help to understand the clinical significance of this bacterium.

Incidence of Penicillium keratitis is low. Five cases of Penicillium were reported by Reddy et al in 1972. [21] A study by Basak, et al from Eastern India reported 10.1 % cases. [22] But a 3-year study from south India in 2002 reported only 0.36 % of Penicillium. [17] A study from Kerala reported Penicillium as 4% of microorganisms isolated from CU and 12.2 % of fungal proven culture. [12] A study from south Kerala showed 20.09% Penicillium in 2011. [13] There are not many cases reported from central Kerala. [11] There is a wide variation in incidence of Penicillium CU depending on geographical distribution, climate, and seasonal variation. Usually, Penicillium is reported in immunocompromised states like HIV, DM. We report a case in an immunocompetent patient from central Kerala. Similar case in an immunocompetent patient was reported from Maharashtra in 2014. [23] KOH showed numerous septate fungal hyphae. LPCB shows chains of single cell conidia borne on phialides with a brush like appearance.

Thanathanee et al reported the second case of keratitis caused by Chrysosporium in 2017 which was successfully treated with Voriconazole. [24] We followed the same treatment with topical and intra stromal Voriconazole and became successful in healing the ulcer. First case of Chrysosporium parvum keratomycosis was reported by MD Wagoner et al in 1999. [25] It was not responsive to Natamycin or Amphotericin B. First case needed PKP as the ulcer perforated. It was a keratinophilic filamentous fungus isolated from soil. [24] Here the ulcer started after exposure to mud. Although human infection is very rare, it can lead to serious systemic or ocular diseases. It presented as numerous filamentous septate hyphae with apical or intercalary spores. Colonies are granular, flat white cream, tan to pale brown. Voriconazole can be used in drug resistant fungal keratitis or rare fungal species causing keratitis. [26] Final culture report was sterile. Standardised susceptibility testing methods using ocular strain may be required for future decision making and management. Clinical correlation is the most important step.

Typical clinical picture of Nocardia keratitis is a well-defined epithelial defect with scalloped margins, white granular appearance, discrete yellowish white pin head sized infiltrates. Stromal infiltrate has feathery margins and a wreath pattern with satellite lesion and associated deep corneal suppuration and vascularisation. Gram stain shows pus cells and a single bunch of beaded, branching thin long gram-positive filaments. Modified Ziehl Neelsen (1%H2SO4) shows acid fast bacilli. Culture shows chalky white colonies. A study by Bharathi, et al in 2003 reported 1.42% Nocardial keratitis. [27] Another ten-year study from south India by Prajna, et al in 2015 showed 1.6 % Nocardia CU. [28] Meera et al had reported a similar case in 2021 from Kerala. [16]

CU caused by dematiaceous fungi typically presents with dry thick raised corneal surface, stromal infiltrate with feathery margins, typical satellite lesions, dendritic pattern, white immune ring in mid periphery, hypopyon and deep stromal infiltration. Healing response and vision are good. Here our case also presented with initial dendritic picture with stromal infiltrate with feathery margins and endothelial plaque with hypopyon and healed with good final visual acuity. Initially we suspected viral aetiology due to this dendritic pattern. Microscopy showed pigmented hyphae and conidiophores with 3 to 6 Conidia that were cylindrical to sausage shaped. Culture showed slow growing suede like with radial furrows, whitish grey to olivaceous grey. First report of a new corneal pathogen Phaeoacremonium parasiticum was reported in 2020 by Horace Massa et al. [29] It was first reported in 1974 as a cause for endophthalmitis in immunocompromised patient. Out of the 34-44% fungi, in tropics 8-17 % of keratitis caused by dematiaceous fungi including Curvularia, and among the dematiaceous fungi, Acremonium species accounts for 2.4%. [30]

One case of successful salvage of eye due to Cylindrocarpon lichenicola keratitis is reported from Kerala in 2020. [15] Another case of Cylindrocarpon is reported from Tamilnadu by Kaliamuthu et al in 2006. [31] Another case of fungal keratitis caused by Cylindrocarpon was reported from Europe by Ganjoux et al in 2012. [32] All these eyes needed PKP to save eyes along with Voriconazole and Natamycin. Microscopy showed branched septate hyaline hyphae. Cylindrical to fusiform, hyaline macroconidia each with 3 to 6 septae with rounded apex were seen. Culture showed white floccose aerial mycelium that turns pale brown.

Curvularia is a dematiaceous filamentous fungus with septate brown hyphae and multiseptated conidia. Colonies are white to pinkish grey initially and turns olive brown or black on maturing. Risk factors include trauma with plants or dirt. Typically, ulcer presents with superficial feathery infiltrate of central cornea to suppurative ulceration. Hypopyon is rare and indicates risk of complication. Dematiaceous fungi are uncommon, but in tropics it causes 8-17 % keratitis. Of all the dematiaceous fungi 66.3% are Curvularia. [30] A 10-year review study done in south India by Gopinathan et al shows 2.8 %

Curvularia among fungal keratitis aetiology in 2002. [33] Another 3-year study in south India reports 2.64% Curvularia keratitis. [22] A study from central Kerala reports equal incidence of Curvularia and Aspergillus (13.04%). [11] No other study from Kerala, or south India reports such higher incidence of Curvularia. [10, 12,13] the higher incidence may be due to the geographical and seasonal variations shown by the fungal etiological agents.

It is very difficult to culture rare organisms and specially to subculture from the culture. Final result may be got as sterile as mixed growth or contamination can occur on subculture. Hence follow up of the culture sent has to be done meticulously so that we don't miss the organism and always correlate clinically. Organisms grown on VITEK is not taken into consideration by many especially if pathogenic organisms are rare and hence not reported. But literature says some organisms may be detected by VITEK only and not easy to culture. Modern diagnostic techniques have evolved and by using them we may be able to diagnose many more pathogenic organisms causing CU much early and which we are missing now by the present culture method. Identifying specific organism early is necessary for starting specific treatment early thus preventing complications and corneal blindness.

Conclusion

This case series highlights the importance of suspecting and diagnosing uncommon pathogens causing CU regionally and treating them appropriately and the need for further studies in this regard.

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