

Original research article

Epidemiological and Microbiological trends of Mycotic Keratitis in South India.

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Abstract

Background: Corneal blindness is a major public health problem globally and infectious keratitis tend to be a predominant cause. Fungal corneal ulcer found to be very common in India due to the tropical climate and a large agrarian population that is at risk. The aim of the study is to identify the risk factors, clinical characteristics, causative fungal agent and treatment outcome of fungal keratitis in South India.

Methods: A prospective study was conducted in all patients with clinically suspected infectious keratitis at Chalmeda Anand Rao Institute of Medical Sciences, Karimnagar, Telangana from January 2019 to December 2021 to determine the causative fungal agents and to identify the risk factors of fungal keratitis. Direct microscopy of corneal scrapings was done by Gram's staining and 10% KOH preparation Fungal culture was done using SDA medium and incubated at 25°C and 37°C. Fungal culture reports were declared negative at the end of 14 days.

Results: Of the 358 samples processed, 224 were identified as infectious keratitis, bacterial organisms were identified in 98 (27.37%) cases and fungal isolates were identified in 126 (35.19%) cases, of which 17 (13.49%) cases were mixed with bacteria and no organism in 134 (37.43%). This study reports mycotic keratitis was predominantly due to the filamentous fungi, *Aspergillus spp.* and *Fusarium spp.* and trauma was to be most common predisposing risk factor observed.

Conclusion: Fungal keratitis continues to be an important cause of ocular morbidity, a strong clinical suspicion of fungal keratitis is crucial in early diagnosis. Direct microscopic detection of fungal structures in corneal scrapings permits a rapid presumptive diagnostic tool in timely and specific management. By choosing appropriate antifungal agents we can combat the disease efficiently, and prevent blindness.

Key words: Corneal infections, Fungal keratitis, *Aspergillus spp.*, *Fusarium*, Antifungal agents.

Introduction

Cornea is the major refractive and protective outer layer of eye, inflammation of the cornea is known as keratitis. There are several causes of keratitis, infectious, physical or chemical.¹ Many microorganisms like bacteria, virus, protozoa, chlamydia and fungi can cause keratitis leading to corneal ulcers.² Bacterial keratitis is the most prevalent amongst microbial keratitis.¹ Causative organisms include bacteria, viruses, fungi and protozoa. Fungal keratitis (FK) was first documented in 1879, and its incidence has been increasing for the past 30 years.³ In tropical regions, fungal corneal ulcers may account for more than half of reported cases, so a key distinction for management is whether the cause is bacterial or fungal.⁴ Microbial keratitis has long been a challenge for the physicians' due to its varied presentation, overlapping

symptoms, and rapid progression. Bacterial keratitis is the most prevalent amongst microbial keratitis.¹

Mycotic keratitis commonly known as fungal keratitis (FK), accounts for approximately 1–44% of all cases of microbial keratitis, depending upon the geographic location.⁵ Mycotic keratitis is a severe and potentially blinding infection of the cornea, especially in tropical and subtropical countries and is considered an ophthalmic emergency. Infected individuals are frequently young agricultural workers of low socioeconomic status. It is one of the leading causes of microbial keratitis (MK) or corneal ulcer and prognosis is markedly worse compared to bacterial keratitis.^{5,6} The most common risk factors ascribed for mycotic keratitis are trauma with contaminated objects with soil, vegetative matter contaminated contact lenses, occlusion of lacrimal duct, fungal skin infections, long term usage of corticosteroids or antibiotics locally or systemically, others include history of eye surgery and eye lid abnormalities.^{2,7}

Fungal keratitis is caused by yeasts and filamentous fungi. A total of 105 species of fungi classified in 56 genera have been identified as the etiological agents of fungal Keratitis.⁸ Filamentous fungi, such as *Fusarium* and *Aspergillus*, and yeast-like fungi, such as *Candida*, are most commonly associated with keratitis. Many other species have also been frequently reported, ranging from *Curvularia* and other phaeohyphomycetes, *Scedosporium apiospermum* and *Paecilomyces*.⁸

In India, *Fusarium* and *Aspergillus* species are being isolated from corneal ulcers in large numbers, irrespective of the geographical area followed by *Curvularia*, *Bipolaris*, and *Candida*.⁵ The causative agents of infectious keratitis vary regionally, with fungal organisms more common in tropical climates and bacterial organisms more common in temperate climates. India is a tropical country that accounts for a large portion of the global burden of infectious keratitis.⁹ The importance of fungal keratitis gained momentum in 2005 following the outbreak of fungal keratitis among contact lens wearers in many developed countries.¹⁰

Fungal keratitis, traditionally manifests as a suppurative lesion with a dry, raised ulcer with crenate, speculated or pseudohyphate borders, satellite lesions and hypopyon; associated with failure to respond to antibacterial treatment. Early and accurate diagnosis is crucial in allowing specific treatment in time, which remains the main stay of vision saving management.⁶ Nevertheless, rapid and intensive empirical treatment is essential till the time of microbiological diagnosis is initiated as an emergency procedure. To institute an appropriate empirical therapy, awareness on clinicoepidemiological pattern of mycotic keratitis is essential.¹¹

Materials and methods

This prospective study was conducted in the department of Microbiology, Chalmeda Anand Rao Institute of Medical Sciences, Karimnagar from January - 2019 to December - 2021. The study was approved by institutional ethics committee. Diagnosis of fungal keratitis was established in 60 non repetitive cases fulfilling the inclusion and exclusion criteria was included in the study. Corneal scrapings obtained from all cases of keratitis, were investigated by direct microscopy with potassium hydroxide 10%, and inoculated for fungal culture.

a. Inclusion criteria:¹²

1. A fungal corneal ulcer classically presents as a dry, raised lesion with crenate or feathery borders, presence of satellite lesions and a hypopyon.
2. All cases with clinical features suggestive of fungal keratitis such as, raised, ulcer/plaque with crenated or feathery margins, dry surface, presence of satellite lesions, weiss ring, endothelial plaque, colouration of the ulcer other than yellow (suggestive of diatomaceous fungi) and thick hypopyon were noted and categorised as patients with fungal keratitis.
3. Patients with corneal perforation
4. Patients whose corneal infection not cured or infection progressing with natamycin 5% eye drops used for atleast 1 week
5. KOH positive keratitis cases.

b. Exclusion criteria:¹²

1. Non-suppurative keratitis such as viral keratitis.
2. Neurotrophic ulcers.
3. Patients with signs and symptoms of endophthalmitis & panophthalmitis
4. Sterile neurotrophic ulcers and any ulcer associated with systemic autoimmune conditions.
5. Degenerative causes were excluded as Mooren's ulcer.
6. Interstitial keratitis,

After selection of patients, standardised proforma was filled for each patient documenting, age, sex, domicile, education, economic status, occupation and predisposing factors including history of trauma, diabetes mellitus and operation (if any). Detailed clinical history was taken and any previous usage of medications recorded. Gram's staining, 10% KOH preparation, bacterial culture using blood agar and chocolate agar. Fungal culture was done using Sabouraud's dextrose agar medium and incubated at 25°C and 37°C. Fungal culture reports were declared negative at the end of 14 days. Dematiaceous fungi were identified by their colony characteristics on sabouraud dextrose agar and by the morphological appearance of the spores in lactophenol cotton blue stain and, in some cases, by slide culture method.

Results

Over the two years of study from January 2019 to December 2021, of the 358 corneal ulcer patients presumed with infectious keratitis, corneal scrapings were collected and subjected for microbial investigation, as direct microscopy with KOH mount and fungal culture. Overall, 224 of the 358 cases presented as infectious keratitis, fungal isolates were identified in 126 (35.19%) cases and bacterial organisms identified in 98 (27.37%) cases, 17(13.49%) cases mixed with bacteria and no organism in 134 (37.43%).

Overall fungal positivity was seen in 126 (35.19%), mycotic keratitis by culture were diagnosed in 118 (93.65%) cases and 8 (6.34%) by potassium hydroxide preparation. Males 73(57.93%) were more commonly affected than females 53 (42.06%). Maximum number of samples 46 (36.50%) were received from age group of 41-50 (Table I). Majority of the patients are from rural area and 74 (58.73%) of patients are farmer by profession, followed by labourer 23(18.25%), home maker 12 (9.52%), tradesman 7(5.55%), carpenter 5(3.96%), and others (3.96%). (Table II). Corneal trauma was identified as the major risk factor predisposing for mycotic keratitis, In our study highest number of cases due to trauma with vegetative matter 81 (64.28%) (Table III). Most common species was *Aspergillus* species 53 (42.06%) followed by *Fusarium* 45 (35.71%) (Table: IV).

Table I: Age wise distribution

| Age in years | No. of cases | Percentage |
|--------------|--------------|-------------|
| <20 | 2 | 1.58% |
| 21-30 | 5 | 3.96% |
| 31-40 | 23 | 18.25% |
| 41-50 | 46 | 36.50% |
| 51-60 | 37 | 29.36% |
| 61-70 | 13 | 10.31% |
| Total | 126 | 100% |

Table II: Distribution according to occupation

| Occupation | No. of cases | Percentage |
|-----------------|--------------|-------------|
| Farmer | 74 | 58.73% |
| Manual labourer | 23 | 18.25% |
| Carpenter | 5 | 3.96% |
| Home maker | 12 | 9.52% |
| Tradesman | 7 | 5.55% |
| Others | 5 | 3.96% |
| Total | 126 | 100% |

Table III: Distribution according to predisposing risk factors

| Risk factor | No. of cases | Percentage |
|------------------------------|--------------|-------------|
| Trauma with vegetative mater | 81 | 64.28% |
| Trauma with stone | 3 | 2.38% |
| Steroids and antibiotics | 9 | 7.14% |
| Wearing of contact lens | 11 | 8.73% |
| Ocular surgery | 14 | 11.11% |
| Others | 8 | 6.34% |
| Total | 126 | 100% |

Table IV: Distribution of fungal isolates.

| Organism isolated | No. of cases | Percentage |
|-------------------|--------------|-------------|
| Aspergillus spp. | 53 | 42.06% |
| Fusarium | 45 | 35.71% |
| Rhizopus | 13 | 10.31% |
| Candida | 6 | 4.76% |
| Cladosporium | 1 | 0.79% |
| No growth | 8 | 6.34% |
| Total | 126 | 100% |

Discussion

Blindness due to corneal infections is a serious problem next to cataract and fungal keratitis is one of the major causes of visual loss globally. According to the World Health Organization report, it is estimated that ocular trauma and corneal ulceration result in 1.5 to 2 million new patients of corneal blindness annually. In India, the incidence of corneal ulceration is reported to be 1130 per million population.¹³ Our study showed a high incidence of fungal isolates 35.19% than bacterial isolates 27.37%, which was similar to the findings of Pratik Narendra Mohod *et al.*,¹⁴ and Basak *et al.*¹⁵ A decline in bacterial keratitis was also found at

several tertiary care centres in south India, due to usage of antibiotics, an increased access to antibiotic eye drops may result in earlier successful treatment of bacterial keratitis.

This study showed significant male preponderance in fungal keratitis, related to majority of studies.^{10,12} In contrast a study by Tanure MA *et al.*,¹⁶ from Philadelphia reported slightly female predominance and 41-50 years is the most commonly affected age group in both sexes in our study similar.^{8,17} In any region, the trends varies with different age groups and different seasons of the year. India is a country with various geographical features and atmospheric conditions, diverse seasonal deviations and disparate flora and fauna in different regions.

Direct microscopy of corneal scrapings for fungal elements found to be rapid and highly valuable in diagnosis of mycotic corneal ulcers. The sensitivity of KOH preparation and fungal culture in our study was 96.82% and 93.65% respectively. A study by Palanisamy Manikandan *et al.*,¹⁰ reported 96.1% sensitivity with KOH preparation, Similarly, another study by Bharathi *et al.*¹⁸ reported 99.23% sensitivity. The sensitivity of KOH preparation has been found to be highly valuable.

The sensitivity of fungal culture in our study was 93.65%, whereas a study by Bandyopadhyay *et al.*, reported 72.9% culture positivity. *Aspergillus* spp. was found to be the predominant agent followed by *Fusarium* as the second most common causative agent of mycotic keratitis.^{8,19,20} while other studies reported *Fusarium* as the most common fungal agent isolated from patients with mycotic ulcers.^{9,10,14,17,21} India being a tropical agricultural country with seasonal increase in temperature and humidity, has a higher prevalence of fungal keratitis. The predominant antecedent factor was trauma with vegetative matter 81(64.28%) followed by ocular surgery and usage of steroids similar findings were observed in other Indian studies.^{8,12,22}

Fungal keratitis is difficult to treat and may result in worse outcomes than bacterial keratitis. There is a major depletion in ocular morbidity with rapid diagnosis and timely treatment. Delay in diagnosis, corneal fungal infections, may result in irreversible sequelae.

Conclusion

This study emphasizes that the relative prevalence of fungal keratitis is high compared to other parts of India, the proportion of infectious corneal ulcers with bacterial pathogens has decreased significantly. Most common risk factors include vegetative trauma, usage of contact lens use, prolonged non-judiciary steroids prescription. The predominant genera of fungi involved were *Aspergillus* and *Fusarium*, patients who fail to respond to medical therapy required surgical intervention, including therapeutic penetrating and lamellar keratoplasty. Therapeutic keratoplasty continues to remain an important treatment modality. Newer antifungals with greater penetration compared to natamycin have shown promising results in the treatment of mycotic keratitis

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