Original Research

MICROBIAL PROFILE AND PREDISPOSING FACTORS OF MYCOTIC KERATITIS IN A TEACHING HOSPITAL, CENTRAL INDIA

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ABSTRACT

Introduction: Mycotic keratitis is one of the commonest causes of eye infections especially in male which works mainly in outdoor. Filamentous fungi (mould-like fungi) were the common cause of Mycotic keratitis and common presentations seen in developing countries. The aim of present study assess predisposing factors and microbial profile of Mycotic keratitis in central India. **Methods:** The study was conducted in the collaboration of Department of Microbiology and Ophthalmology in G. R. Medical College, Gwalior, India. Samples were collected from corneal scraping by sterile cotton swab and inoculated on culture media Sabouraud dextrose agar (SDA) for fungal culture. Material obtained from corneal scrapping also performed 10% KOH mount preparation, gram's staining and Lactophenol cotton blue (LPCB) mount. Identification of fungal isolates was done by colony morphology, growth characteristics and relevant standard tests. **Results:** A total of 237 out of 740 (32%) fungal isolates was obtained from suspected patients of Mycotic keratitis, out of which *Fusarium* species (33.2%) was the most predominant followed by *Curvularia* species (21.9%) and *Aspergillus* species (16.7%), Ocular trauma was the key predisposing factors. **Conclusion**: Trauma was the key predisposing factor of keratitis because of filamentous fungi. The commonest causative agent of fungal keratitis in current study was *Fusarium* species followed by *Curvularia* species. Early proper diagnosis and management of fungal keratitis reduces the ophthalmic morbidity and mortality.

Keywords: Culture, filamentous fungi, fungal keratitis. Keratomycosis

Introduction:

Microbial keratitis is a leading cause of ocular morbidity and mortality in developing countries, secondary to the cataract, (Flaxman et al., 2017). Etiology of the microbial keratitis includes bacterial agents, viruses, and fungal agents, amongst them Mycotic keratitis cause more serious corneal infection may lead to blindness. It still remains the challenging issue of all ophthalmologists (Khor et al., 2018 and Acharya et al., 2017).

Incidence of Mycotic keratitis was very according to geographical locations, population of patients, corneal health, mainly occurs in summer seasons, tropical climates and more among agricultural workers due to vegetative trauma (Estopinal et al., 2016). Hence, knowledge of risk factors, causative agents and epidemiological characteristics features in particular areas are critically useful in early recognition, prompt therapy, proper management and prevention of the microbial keratitis. Proper laboratory investigation was necessary before the starting specific therapy. For the diagnosis of Mycotic keratitis culture of corneal scrapings on SDA media and microscopy were necessary for identification of the etiological agent (Bashir et al., 2005). Mycotic keratitis is leading cause of blindness, because it difficult to diagnosed, cause more serious infection, poor prognosis, less responding to treatment and worse visual outcome (Prajna et al., 2013). Etiological agents of Mycotic keratitis were yeast like fungus and filamentous fungi, yeast like fungi are *Candida* species was prevalent in temperate climates and filamentous fungi are *Aspergillus* spp. & *Fusarium* spp. were prevalent in tropical climates (Shah et al., 2011 and Galarreta et al., 2007).

The objective of the study was to detect the predisposing factors and isolation of fungal agent from corneal scrapping for diagnosis of fungal keratitis prevalent in central Indian populations.

Methods:

A two-year prospective study of fungal keratitis was carried out from July 2016 to June 2018. Only patients attending the ophthalmology clinic and were with microbiologically proven fungal keratitis were enrolled in our study. Detailed history was obtained with socio-demographic and occupation data, ophthalmic medical and surgical history, contact lenses and immunosuppression history.

The corneal scrapping material was collected by Ophthalmologists by using strict sterile precaution under slit-lamp examination.

Sample was collected from base and edges were subjected to microscopic examination using freshly prepared 10% KOH and Gram staining, (ICMR 2nd edition., 2019) and, were inoculated on two plates/tubes of Sabouraud's dextrose agar media with antibiotics incubated at 25^{O}C and 37^{O}C separately for 4 weeks. The corneal scrapping was also cultured on blood agar media plates in the pattern of "S" or "C" shaped streaking, to ensure the growth in inoculum from rather than contaminant from the lab. All inoculated culture Medias was incubated aerobically. In the first week time cultures plates were checked daily whereas on remaining next three weeks duration checked twice a week.
Identification of fungal agents was done by standard laboratory methods via microscopy, characteristics features of growth; colony morphology, according to ICMR 2nd edition (2019), Lactophenol cotton blue (LPCB) preparation and slide culture.

Statistical analysis: statistical analysis was performed by SPSS version 22 software. Percentages and mean value were calculated and analysed. P< 0.05 considered statistically significant

Ethical consideration: The present study was ethically approved by institutional board of ethical committee; we have obtained informed consent from all the study participants

Results:

During the study period, out of 740 patients, 237 (32%) patients were either smear or culture or both positive Mycotic keratitis cases were enrolled in our study. Out of 237 cases, 197 (83.12%) were both positive (KOH and Culture), 27 (11.40%) KOH only positive, 13 (5.48 %) culture only positive and those were negative for both, were excluded from the study group. Comparison of KOH mount and culture isolates were shown in table 1.
Current study found the rate of fungal keratitis was reported higher among males 142 (59.92%) than female 95 (40.08%). Most of the patients belong to the age group of 41-60 years. The common associated predisposing factors in descending order includes trauma from vegetative objects 53%, contact lens user 31%, on steroid therapy 29%, diabetic and hypertensive 27%, previous ocular surgery 23% and ocular surface disorder 21% as shown in figure:1.
Among the 237 mycological proven cases of Mycotic keratitis the commonest fungal isolates obtained was *Fusarium* species 33.2% (n=79) followed by *Curvularia* species 21.9% (n=52). Detail description of microbial isolates were shown in Figure 2
It proves that KOH is much more sensitive than culture in picking up fungal infections, and can save valuable time on diagnosis & treatment.

Table 1: Comparison of KOH mount and culture of fungal isolates

Statistic	KOH Mount	Culture
Sensitivity	93.81%	89.65% to 96.66%
Specificity	0.00%	0.00% to 12.77%
Positive Likelihood Ratio	0.94	0.91 to 0.97
Disease prevalence	88.61%	83.86% to 92.36%
Positive Predictive Value	87.95%	87.57% to 88.31%
Negative Predictive Value	0	--
Accuracy	83.12%	77.73% to 87.66%

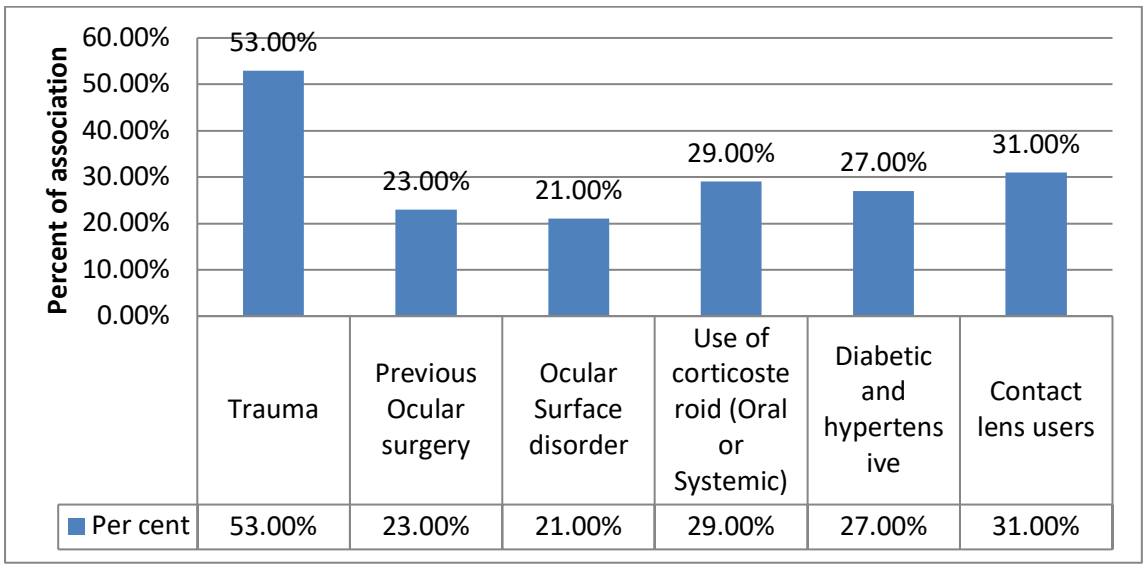

Figure1: Distribution of associated predisposing factors

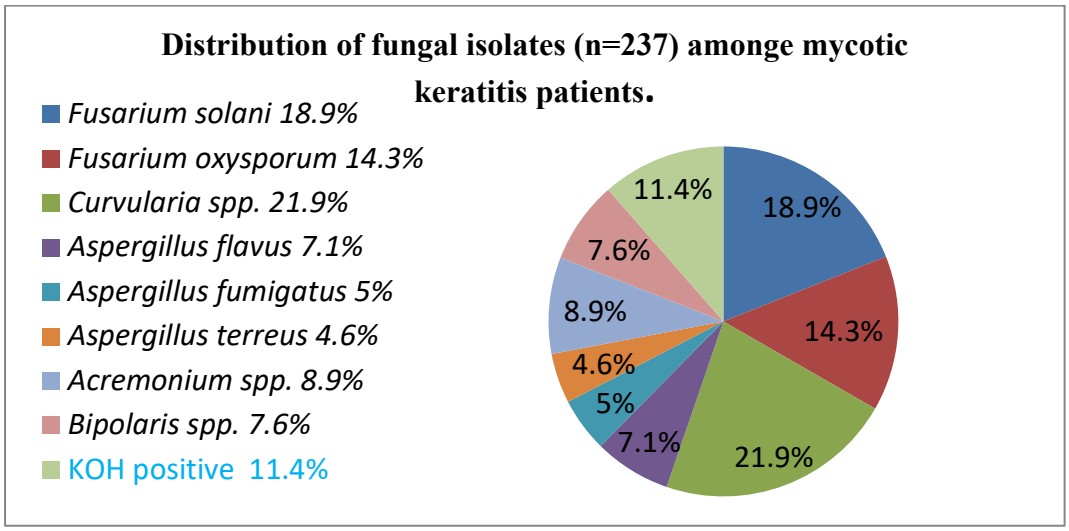

Figure 2: Distribution of fungal isolates among Mycotic keratitis patients

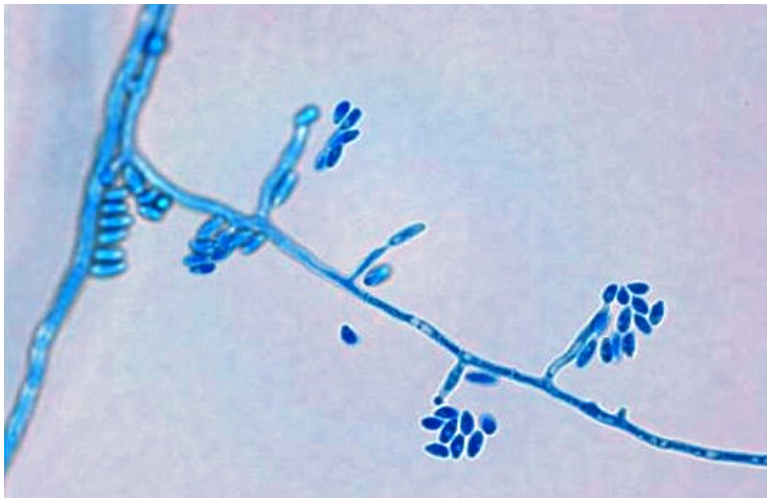

Image 1: LPCB mount shows Fusarium Solani

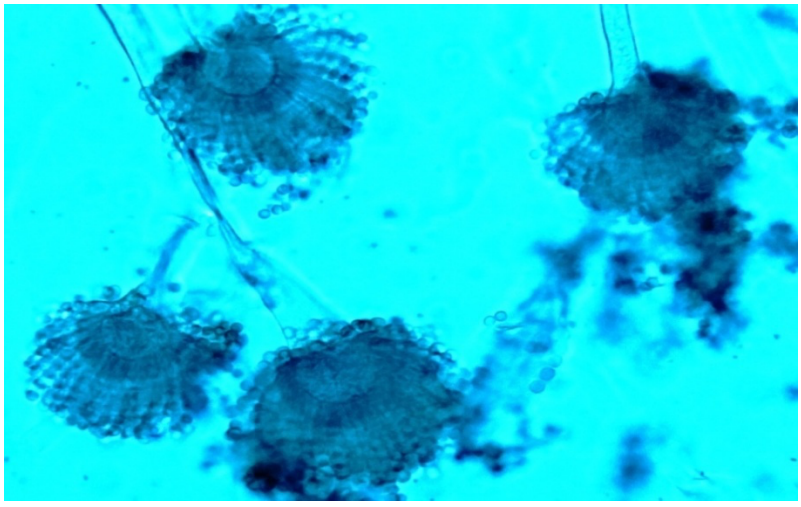

Image 2: LPCB mount shows Aspergillus flavus

Discussion

The Mycotic keratitis is a leading cause of ocular morbidity and mortality. In India and other tropical countries fungal infection of eye is most frequently encountered condition.

Incidence of Mycotic keratitis in current study was 32%, Similar incidence was also reported by many other studies like, (Kalshetti et al., 2015, Rautaraya et al., 2011, Jiragyal et al*.,* 2016 and Tewari et al*.,* 2012), whereas (Puig et al., 2020) reported only 7.1% incidence of fungal keratitis, lower than the present study. In contrast to current study (Venkatesh et al., 2018 and Javadi et al., 1996), reported 86% higher incidence of fungal corneal ulcer. These geographical variations may be due to predisposing and etiological agents of Mycotic keratitis was differ in different regions, mainly mycotic keratitis seen in tropical and subtropical region. In the present study the male to female ratio was found to be 1.49, similar results were reported in a study by (Satpathy et al., 2019).

Many studies observed that incidence of Mycotic keratitis was high in farmers (Kunimoto et al., 2000). Farmers are prone to Mycotic keratitis because of more chances of vegetative ocular trauma. In present study higher incidence of Mycotic keratitis observed between ages of 35-60 years. (Kalshetti et al., 2015) cited the higher incidence between ages of 21 to 50 years; this could be due to middle age group male were outside workers having more expose to infectious agents. In the present study, the Mycotic keratitis was due to moulds; predominantly *Fusarium* species followed by C*urvularia* & *Aspergillus* species, our finding was comparable with the (Bharati et al., 2003 and Chitamparam et al., 2020) whereas (Chander et al., 2008)*,* reported *Aspergillus* species was the most common isolate in their study

Generally, most of the fungal agents do not invade the intact corneal epithelium, penetration of agents made after trauma (animal or plant agent). Trauma can facilitate the implantation of fungal conidia in corneal stroma or indirect abrasion of corneal epithelium permit fungal invasion (Thomas et al., 2013).

In the present study, Ocular trauma is the key predisposing factor, occurring in 53% of patients, comparable with the other studies like, (Waghmare et al., 2019, Singh et al., 2020 and Hoffman et al., 2021) Whereas (Menard et al., 2022) reported contact lens was the most common risk factor of fungal keratitis.

Conclusion:

Incidence of Mycotic keratitis was 32%, more in male than female in the present study. Ocular trauma was the key predisposing factor of fungal keratitis. Fusarium, Curvularia and Aspergillus were the most common fungal agents isolated from Mycotic keratitis patients. The fundamental element in the diagnosis of Mycotic keratitis is clinical suspicion by an ophthalmologist along with laboratory diagnosis by direct microscopic examination and culture. Early definitive diagnosis of Mycotic keratitis by culture may help in proper management.

Conflicts of Interest

The author declares no conflicts of interest.

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