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Identification of Fungal Species in Proved Cases of Fungal Corneal Ulcer

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ABSTRACT

Background: *Mycotic keratitis is a fungal infection of the cornea. This infection is difficult to treat and it can lead to severe visual impairment or blindness. It is worldwide in distribution, but is more common in the tropics and subtropical regions. Trauma is the major predisposing factor, followed by ocular and systemic defects, prior application of corticosteroids. The objective of this study was to determine causative agents of mycotic keratitis.*

Methods: *A retrospective review of medical and microbiology records was conducted for 25 corneal ulcer patients with suspected fungal etiology who were subjected to direct examination by 10% KOH mount, Gram stain and culture.*

Results: *This study included 25 subjects with corneal ulcers, males were more commonly affected and were mostly in the age group of 45-60 years. Aspergillus was the most common fungus isolated followed by fusarium. The most common predisposing factors were injury to the eye.*

Conclusion: *In this study we found that fungal corneal ulcer is more predominant in males and trauma with vegetative matter is the predisposing factor and we found Aspergillus species is more common.*

Key words: *corneal infections, fungal keratitis, fungal culture, Fusarium spp, Aspergillus spp*

INTRODUCTION

Corneal infection is a leading cause of ocular morbidity and blindness worldwide. Corneal ulceration is a major cause of monocular blindness in developing countries. Surveys in Africa and Asia have confirmed these findings, and a recent report on the causes of blindness world wide consistently lists corneal scarring second only to cataract as the major aetiology of blindness and visual disability in many of the developing nations in Asia, Africa and the Middle East.^[1]

According to the World Health Organization, corneal diseases are among the major causes of vision loss and blindness in the world today, after cataract and glaucoma. In India, it is estimated that there are approximately 6.8 million people who have vision less than 6/60 in at least one eye due to corneal diseases; of these, about a million have bilateral involvement. It is expected that the number of individuals with unilateral corneal blindness in India will increase to 10.6 million by 2020. According to National Programme for Control of Blindness survey, corneal blindness accounts for 0.9%.^[2]

Corneal blindness from healed infected keratitis is one of the most preventable causes of monocular blindness in developing countries.^[3] Almost any microorganism can invade the corneal stroma if the normal corneal defence mechanisms, i.e. lids, tear film and corneal epithelium are compromised. A wide spectrum of microbial organisms can produce corneal infections and consequently the therapeutic strategies may be variable. One of the key elements in this effort is a

proper understanding of the microbiological and clinical characteristics of this disease entity which will enable the ophthalmologist to initiate appropriate antimicrobial therapy.^{[1][3]}

Many micro organisms can cause infectious corneal ulcers. Among them are bacteria, fungi, viruses, protozoa and *Chlamydia*.^[5] Fungal keratitis is caused by invasion of the cornea by filamentous fungi, yeasts, and dimorphic fungi. At least 70 genera of filamentous fungi (56), yeasts (11), and dimorphic fungi (3) have been reported to cause human keratitis.^[4]

Mycotic keratitis is an infection caused by fungus that leads to inflammation and ulceration, usually following trauma or treatment for a bacterial infection with steroids or antibiotics.^[5] Fungal keratitis can cause a deep and severe corneal ulcer. It is caused by *Aspergillus spp.*, *Fusarium spp.*, *Candida spp.*, *Rhizopus*, *Mucor*, and other fungi^[3]. The typical feature of fungal infection is slow onset and gradual progression, where signs are much more than the symptoms. Small satellite lesions around the ulcer are a common feature of fungal keratitis and hypopyon is usually seen. Keratitis due to filamentous fungi is believed to usually occur following trauma, the key predisposing factor, in healthy young males engaged in agricultural or other outdoor work. The traumatising agents can be of plant or animal origin (even dust particles), that either directly implant fungal conidia in the corneal stroma or abrade the epithelium, permitting invasion by exogenous fungi. Environmental factors (humidity,

rainfall, wind) greatly influence the occurrence of fungal keratitis.^[5]

The purpose of this study was to identify the infectious fungal organism causing corneal ulcers seen at a tertiary referral centre K.R hospital, Mysore.

MATERIALS AND METHODS

We conducted a retrospective review of all medical and microbiology records for all cases treated with fungal keratitis. An analysis was performed to study the demographic features of fungi. A detailed history of present illness was undertaken on all patients with special reference to occupation, trauma, medication to eye and surgical intervention, immunosuppression, and use of cosmetic or therapeutic contact lenses.

Following clinical diagnosis of infective corneal ulcer based on slit-lamp biomicroscopic examination, the patient's corneal were scraped and sent for microbiological investigations as per institutional' protocol. Using standard techniques, corneal scraping was performed to all corneal ulcer patients under aseptic condition using 22 gauge needles or a Kimura spatula following the instillation of local anaesthesia (4% proparacaine eye drops). The material collected from the leading edge and base of the ulcer was inoculated directly onto blood agar, chocolate agar,

McConkey agar for bacteria, and Sabaroud's agar for fungal culture. Two smears were made onto two slides. One slide was stained with Gram stain and the other with 10% KOH preparation for direct microscopic examination. For all cases, empirical treatment was given while waiting for the microbiological test and was later changed according to the results or the patients' responses. Sabouraud dextrose agar were incubated at 27°C, examined daily, and discarded at 3 weeks if no growth was seen.(aet). All culture positive for bacteria are excluded from study.

RESULTS

A total of 25 patients were diagnosed with fungal keratitis from January 2014 to October 2014. Demographic data was summarised in Figure 1 and 2. Males were more common than the females. The majority of patients (48%) were aged between 46 to 60 years old. Four of our patients suffering from fungal keratitis (16%) were diagnosed with diabetes mellitus only during this screening process.

The most common predisposing factors for developing fungal keratitis in our patients was injury to the eye (16/25, 64%) with vegetative matter and preexisting ocular surface disease (dacryocystitis)(8/47, 32%). Two third of the patients who had injuries with vegetative material were directly related to agricultural work.

Fig.1.The Age distribution among study group

AGE(years)	No. of patients	PERCENTAGE
30-45	6	24%
46-60	12	48%
>61 years	7	28%

Fig.2. Sex distribution among study group

SEX	No. of patients	Percentage
Male	17	68%
Female	8	32%

Of the 25 culture positive cases the most frequent agent isolated was *Aspergillus* (60%) followed by *Fusarium*(32%) species

Fig.3. Fungal species among study group

FUNGAL SPECIES	No.of patients	PERCENTAGE
<i>Aspergillus</i> spp	15	60%
<i>Fusarium</i> spp	8	32%
<i>Candida</i> spp	2	8%

DISCUSSION

Filamentous fungi are the commonest cause of mycotic keratitis in many countries in tropical latitudes and it is vital that a specific diagnosis is made as quickly as possible to ensure prompt institution of antifungal therapy. Although a detailed clinical examination may help to reach a rapid presumptive diagnosis, fungal keratitis continues to be confused with other causes of inflammatory keratitis. Certain clinical characteristics of corneal ulcers may suggest a specific pathogen, but it is now generally accepted

that a reliable diagnosis cannot be made by clinical appearance alone and that microbiological investigations should be performed. Unfortunately, many ophthalmologists working in developing countries do not have access to basic ocular microbiological investigations such as microscopy or culture of corneal scrapes. Mycotic keratitis has emerged as a major ophthalmic problem since its recognition in 1879(36) *Fusarium* species was found to be the most common cause of fungal keratitis from south

India (Madurai and Tamilnadu) by Barathi et al. (2002, 2003) and Srinivasan et al. (1997) Gopinathan et al. reviewed the epidemiological features of 1,352 patients with culture-proven mycotic keratitis seen over a period of 10 years at a tertiary care eye hospital in southern India. Males were significantly more frequently affected than females (a ratio of 2.5:1), and 64% of patients were in the age group 16 to 49 years. Ocular trauma was predisposed to infection in 54% of patients; trauma occurred significantly more frequently in those working outdoors than in those who were indoors. Trauma by vegetable material was believed to be a specific risk factor for a fungal infection of the cornea in this series of patients.

Fungal corneal ulcers may be reported at any age and in the present study, the age of the patients varied from 30 to 70 years. However, the most susceptible age group was 45 to 60 years. In addition, fungal corneal ulcers were found to be more common in men than in women. Corneal trauma has been identified as the most common risk factor for mycotic keratitis, which was also the case in the present study. Plant material was reported to be the most frequent traumatizing agent in our series (17 cases).

CONCLUSION

The key element in the diagnosis of mycotic keratitis is the clinical suspicion by ophthalmologists. Fungal corneal ulcer is common in India due to the tropical climate and a large

agrarian population that is at risk. In our study we found out that trauma with vegetative matter is the most predisposing factor in fungal corneal ulcer. Since agriculture sectors contributing to the development of our country and fungal infection are common, improvement in our eye care systems is crucial in preventing blindness in our region.

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