

Keratomycosis - A 15 Year Laboratory Experience

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Keratomycosis is infection of cornea caused by different fungal species. It is most prevalent in agricultural area and hence in India. It is a major blinding disease second only to cataract and accounts for almost 50% of patients with corneal ulcer. This study was done to determine the frequency of occurrence and other demographic characteristics of keratomycosis in and around Davanagere, Karnataka. The present study was done over a period of 15 year from 1996-2011, among patients referred to department of Microbiology, J.J.M Medical college, Davanagere. Corneal scrapings were collected from total of 514 suspected cases of Keratomycosis and investigated. Out of 514 patients with corneal ulcers, 201 were diagnosed as keratomycosis cases (39.1%). Males were most commonly affected (Male : female= 2:1) and seen commonly between the age group 21-40 yrs (70.14%). Most common fungus isolated was *Fusarium* seen in 94 cases (46.76%), followed by *Aspergillus* 41 (20.39%), *Helminthosporium* 17 (8.45%), *Penicillium* ten(4.97%) and *Candida albicans* five(2.48%) etc. Agricultural trauma being the most common predisposing factor accounting 54.72% and agricultural workers are commonly affected group (i.e., 64.67%) in this study.

Key words: Corneal scrapings, Keratomycosis, 10% KOH wet mount, Fungal culture.

According to the WHO, corneal diseases are leading cause of vision loss and blindness, second only to cataract¹. Corneal ulceration is a major cause of monocular blindness in developing countries²⁻³. Approximately 28% of ulcerative keratitis shows fungal etiology, though data varies from 6%-53%⁴.

Yeasts (*Candida* species) are the major cause of keratomycosis in temperate climates whereas, filamentous fungi (*Fusarium* and *Aspergillus*) are the common pathogens in tropical climates⁵.

Keratomycosis is common in our country because of the tropical climate and a large agrarian population at risk. There has been a dramatic increase in the frequency of these infections in the last two decades probably because of indiscriminate use of antibiotics and corticosteroids in ophthalmology practice⁶.

It is important to consider fungi as a etiological agent of keratitis because of the devastating damage it can produce if not diagnosed and treated at the earliest⁷.

So, we conducted this study at the department of microbiology to identify the frequency of occurrence, causative agent and other demographic features of keratomycosis.

MATERIALS AND METHODS

Source of data

Ours is a tertiary care hospital that has input coming from in and around Davanagere

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district. So the present study included corneal ulcer patients referred to the department of Microbiology from ophthalmic OPD of Bapuji hospital, Chigateri general hospital and other ophthalmologists in the district.

Sample size

514 patients with corneal ulcer

Inclusion criteria

- Patients with Corneal ulcer referred to the Department of Microbiology, JJM Medical College, Davanagere.
- Patients on antifungal therapy were also included in the study.
- Consent from Institutional ethical committee has been obtained.

Methodology

A total of 514 suspected cases of Keratomycosis were investigated in the Department of Microbiology. Detailed history of these 514 patients regarding age, sex, occupation, precipitating factors (in terms of injury /foreign body etc) and prior antifungal usage were collected.

Procedure adopted for collecting corneal ulcer scrapings

Corneal scrapings from 514 patients with corneal ulcer were collected and investigated

during period from 1996-2011. Scrapings were collected using sterile disposable needle No.26 after installing 1-2 drops local anaesthetic drug. Laboratory workup included Gram's stain, 10% Potassium Hydroxide (10% KOH) wet preparation, bacterial and fungal culture. For fungal culture, scrapings were inoculated on SDA (Sabouraud's dextrose agar) with chloramphenicol in duplicate (one at 37°C and one at 25°C incubation).

The results of 10% KOH and fungal culture only were considered for making the diagnosis of keratomycosis..

RESULTS

Out of 514 patients with corneal ulcer, fungal etiology was identified in 201 cases (39.1%). This includes cases positive by both KOH mount examination and culture on SDA as well as corneal ulcer cases that were positive by KOH but negative on culture (20 cases). Nine cases that were positive by SDA culture and negative on KOH mount were not included under keratomycosis since contamination could not be ruled out (Table 1). Repetition of scrapings was not possible since patients did not come for follow up.

Table 1. Showing KOH and fungal culture positivity

	KOH +ve SDA Cul +ve	KOH +ve SDA Cul -ve	KOH -ve SDA Cul +ve	KOH -ve SDA Cul -ve	Total
no. of cases	181	20	09	304	514
Percentage(%)	35.21	03.89	01.75	59.15	100

Out of 201 patient's with keratomycosis, males were commonly affected than females (Male : Female = 2:1) and most commonly seen in the age group between 21-40 yrs i.e. 141 cases (70.14%).

Out of 201 cases of keratomycosis, agricultural trauma was the leading predisposing factor 110 (54.72%) compared to other predisposing factors. Foreign body history was seen in 53 cases (26.37%). No predisposing factor was seen in 38 cases (18.91%). (Table 2)

In the present study, keratomycosis is commonly seen in agricultural workers 130 (64.67%), followed by house wives 23 (11.45%), retired/ not working population 14 (6.97%),

students 11(5.48%) wood cutters six (2.98%), teachers six (2.98%), gardeners six (2.98%), flour mill workers five (2.49%). (Fig. 1)

Our study revealed that keratomycosis was commonly seen from June to September than other months in a year.

Table 2. Predisposing factors in patients with keratomycosis

Pre disposing factor	No. of patients
Agricultural trauma	110
Foreign body	53
No history of trauma	38

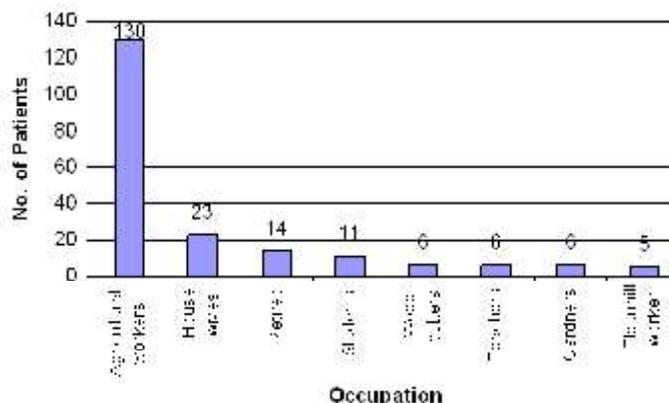


Fig. 1. Occupation of patients with Keratomycosis

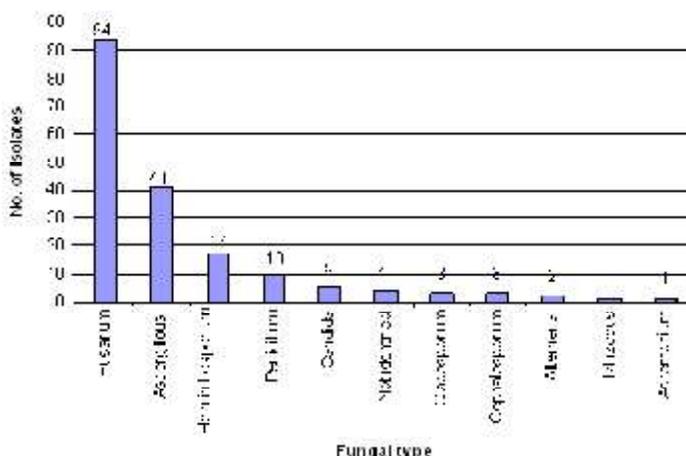


Fig. 2. Spectrum of fungal isolation obtained

In this study, frequently isolated fungi are fusarium spp 94 (46.76%), next common is *Aspergillus* spp seen in 41 cases (20.39%), followed by *Helminthosporium* 17(8.45%), *Penicillium* 10(4.97%), *C.albicans* five(2.48%), three isolates each of *Cladosporium* and *Cephalosporium* (1.49%), alternaria two(0.99%) and single isolate each of acremonium and rhizopus spp (0.49% each) (Fig. 2). Four isolates (1.99%) could not be identified because of lack of sporulation. No growth was seen in 20(9.95%) cases.

DISCUSSION

Mycotic keratitis occurs more frequently in developing countries such as India rather than developed countries^{2,3}. Out of 514 cases with keratitis, 201 cases (i.e., 39.1%) were diagnosed as keratomycosis This includes 181 cases which were

positive by both 10% KOH and fungal culture and 20 cases that were KOH positive and culture negative. The remaining 304 cases did not had fungal etiology.

On culture, 20 yielded no growth inspite of positive KOH mount findings. This may be due to (1) prior antifungal therapy to the patient or (2) inadequacy of the sample for inoculation on SDA. Out of this 20 cases, 16 cases had history of using antifungal eye drops and remaining four patients had no such significant history. These 20 cases were included in the keratomycosis group. Nine cases that were positive by culture but negative by KOH mount were not included due to the possibility of contamination, which could not be ruled out as patients did not come for follow up.

Keratomycosis was found more commonly in men than women⁸⁻¹¹. This may be explained by increased outdoor activities of men

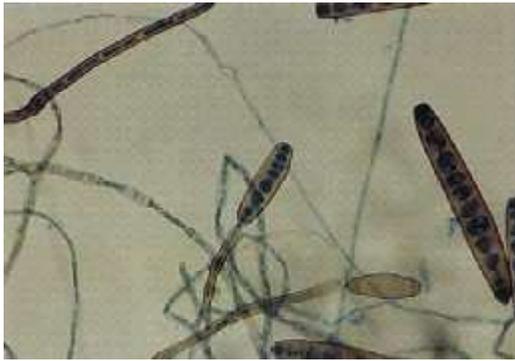


Plate 1. Microscopic appearance of *Helminthosporium*



Plate 2. Colony of *Fusarium* on SDA

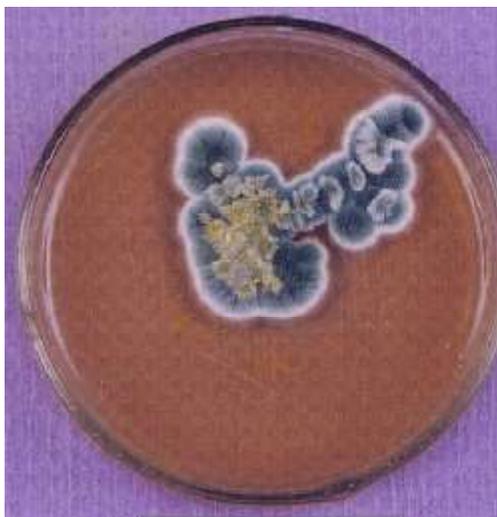


Plate 3. Colony of *Penicillium* on SDA

compared to women and hence greater exposure to fungal agents. The common group affected were in the age group of 21-40 yrs however, corneal ulcer may be seen in any age groups.

Corneal trauma is the most important predisposing factor. In our study, agricultural trauma [includes injury with plant twigs/stick/wood chip/vegetative matter] heads the list of predisposing factors [110 cases (54.72%)] which is similar to other studies ranging from 40-50%^{8,12}. Other predisposing factor was foreign body seen in 53 patients (26.37%) and 38 cases (18.91%) did not show any history of trauma or foreign body.

Agricultural workers are the main victims, they form the major percentage of patients i.e., 130 (64.67%) with keratomycosis.

Environmental factors and seasonal variations such as humidity, rainfall, wind and harvesting may influence the incidence of mycotic keratitis⁸. In our study maximum number of cases were seen from June – September.

A review of literature shows distinct pattern of geographical variation in the aetiology of keratomycosis. In temperate climate, fungal ulcers are uncommon and frequently associated with candida species than filamentous fungi¹³. *Fusarium* and *Aspergillus* spp which are filamentous fungi are the predominant agents reported in tropics¹³.

Fusarium species are commonly isolated from cases of keratomycosis in south India and aspergillus species from north India¹⁴⁻¹⁶. This may be due to change in climatic conditions and other factors. In this study also, *Fusarium* is seen in more number of cases. Among *Aspergillus* species, *Aspergillus flavus* was the most common etiological agent (seen in 24 cases) followed by *Aspergillus fumigatus* (15 cases) and *Aspergillus niger* (2 cases).

Limitation of the study

In the present study corneal ulcer patients who had used local antifungal eye drops were also included. This has probably reduced the sensitivity of culture on SDA.

CONCLUSION

The aetiological agent in ocular infections vary throughout the world. These regional differences in causative organisms of

mycotic keratitis are clinically important because they influence the treatment. In the present study, big proportion of cases i.e. 39.10% has fungal etiology. This could only be representing the tip of iceberg. Hence rapid isolation and identification of the causative fungus is critical in corneal ulcer cases, also there is a need for doing 10% KOH and fungal culture for all cases of corneal ulcer instead of first starting antibiotic therapy randomly and then referring them to microbiology department. In our study mycotic keratitis was seen more in male patients and among agricultural workers. Fusarium was the common isolate. Agricultural trauma was the main contributing factor.

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