



A Clinical Study of Incidence and Morbidity Pattern of Fungal Keratitis With or without Bacterial Infection

KEYWORDS

Suppurative keratitis, Mycotic keratitis, Ocular trauma

Dr. R. C Paul

Assistant Professor, Department of Ophthalmology, Tripura Medical College & Dr. B.R.A.M Teaching Hospital, Tripura(W)-799014.

*** Dr. A. K. Chakma**

Professor, Department of Ophthalmology, Tripura Medical College & Dr. B.R.A.M Teaching Hospital, Tripura(W)-799014.
* Corresponding Author

Dr. D. Datta

Professor & HOD, Department of Ophthalmology, Tripura Medical College & Dr. B.R.A.M Teaching Hospital, Tripura(W)-799014.

ABSTRACT **Aim :** To study the incidence and morbidity pattern of fungal keratitis with or without bacterial infection in Tripura. **Materials and Methods:** In this study 300 numbers of corneal scrapings were collected from patients presented in the department of ophthalmology since 1/11/2013 to 31/10/2015, with the clinical diagnosis of corneal ulcer with or without hypopyon were subjected for microbial investigation to find out the etiological agents. **Result & observation:** Out of 300 cases 180(60%) were found to be culture positive, of this 102 (56.6%) patients had bacterial growth, 78 (43.3%) had fungal growth and 19 cases had mixed bacterial and fungal growth. The Males are the more preponderant sex group accounts 68%. Staphylococcus aureus was the most commonly isolated bacterial organism (23.8 %) followed by Staphylococcus epidermidis (11.67%), Escherichiae coli(7.77%), Pseudomonas spp (6.11%), Klebsiella sp(5%) and Acinetobacter spp(2.2%) the most frequently occurring gram-negative organism. Aspergillus spp (Aspergillus flavus, fumigatus & niger) were the major fungal etiological accounts 16.15% of the total 78 (43.3 %) fungal corneal ulcers followed by Candida albicans(11.1%), Fusarium solani(6.67%), Penicillium species(3.3%), Rhodotorula spp(1.16%) & Trichosporon spp(1.1%). **Conclusion:** Mycotic keratitis will continue to be an important cause for ocular morbidity despite of bacterial agent. Clinical suspicion by the ophthalmologists should be confirmed by microbiological finding prior to initiating corticosteroids and anti-bacterial antibiotics. Precise identification of the causative fungus and institution of appropriate treatment strategy could save the blindness of the eye.

Introduction:

Corneal blindness is a major public health problem worldwide and infectious keratitis is one of the predominant causes for blindness followed by cataract especially in developing countries in comparison to developed countries where 90% people live with this problem (WHO). worldwide 161 million people are visually impaired, 124 millions low vision and 37 millions blind, but the magnitude is much more in India, having one-fourth the world's total blind population¹⁻³. As per recent study Corneal scarring second only to cataract as the major etiology of blindness and visual disability in many of the developing nations in Asia, Africa and the Middle East⁴. A wide spectrum of microbial organisms can produce corneal infections by invading the corneal stroma when defence mechanism of corneal epithelium are compromised⁵. The incidence of mycotic keratitis is more in tropical and subtropical countries due to the climatic environment. Impact of climatic changes by altering local weather or ecological patterns effect human health by resulting bacterial and fungal proliferation^{6,7}. Climatic conditions of North Eastern part of India prevailing with heavy rainfall and humidity throughout the year may result proliferation of microbes in traumatised eye. Most of the people are living under poverty line; hence cultivation, farming, intensive labouring etc are being the only profession for their survival. These may predispose the reason for traumatization of the cornea accidentally or incidentally. However there is always a poor data available in this region which practically emphasised design the present study.

Materials and Methods:

After a thorough clinical evaluation and slit lamp biomicroscopic examination by a dedicated ophthalmologist samples collected from all the 300 suspected patient of

corneal ulcer who were attending in OPD of ophthalmology department of Tripura Medical College during last two year. The Ulceration and Suppurative keratitis was defined as a loss of the corneal epithelium with underlying stromal infiltration and suppuration associated with signs of inflammation with or without hypopyon. Typical viral ulcers and healing ulcers were excluded as were Mooren's ulcers, marginal ulcers, interstitial keratitis, sterile neurotrophic ulcers, and any ulcers associated with autoimmune conditions. All the Clinically suspected patients with infectious keratitis of different of age group except preschool children in different sex were included in our study. Once a clinical diagnosis of corneal ulcer was made, their Socio-demographic data, duration of symptoms, predisposing risk factors, clinical details, prior treatment modalities and associated ocular and systemic diseases etc are also recorded in a standardized patient data sheet pro-forma (Table-1).

Table No.01: Standard Clinical Examination Form for Corneal Ulcer Patient Proforma

Patient details	Patient number:
Name: _____	Sex: M F
Age: _____	
Address: _____	
Ophthalmic History:	
Trauma _____	Eye surgery _____
Ocular surface disorder _____	Contact lens wears _____
Corneal exposure _____	Trichiasis _____
Diabetes mellitus _____	Dacryocystitis _____

Does the patient have a history of diabetes? Y/N.
If yes, for how long _____

If other, give details:
 Current topical antibiotic Y / N specify _____
 Current topical antifungal Y / N specify _____
 Current topical steroid Y / N specify _____
 Traditional eye medicine Y / N specify _____
 Presentation Date of primary presentation ___/___/___
 Eye RE / LE / Bilateral
 Duration of symptoms _____ days
 Visual Acuity (uncorrected) Right _____ Left _____

BASE - LINE EXAMINATION:

Ulcer size: _____ **Diagram:** _____

Lid Edema: Mild/Moderate/ severe

Depth of Ulcer: Deep/Superficial

Depth of infiltrate:Anterior stroma /Mid stroma/ Posterior stroma

Hypopyon: Absent/ Present

Height _____mm

REVIEW Date: ___/___/___

MICROBIOLOGY RESULTS

Gram stain _____ date ___/___/___

KOH _____ date ___/___/___

Locto phenol

Cotton Blue _____date ___/___/___

Culture (BA) _____date ___/___/___

Culture (SDA) _____date ___/___/___

Assessment:

Eye: RE / LE / Bilateral

Visual Acuity Right _____ Left _____

Diagram with ulcer size

Depth of infiltrate: Mid stroma/Anterior stroma/Posterior stroma

Hypopyon: Absent/ Present

Height _____mm

Healing/Working/No Change/Perforation

After a detailed ocular examination of every patient corneal scrapings were obtained under magnification of slit lamp or operating microscope after instillation of 4% lignocaine (lidocaine) without preservative by using standard techniques, by an ophthalmologist under aseptic conditions from each ulcer using a sterile Bard-Parker blade (No 15). The scraping material were obtained from the leading edge and the base of each ulcer directly onto sterile slides for 10% KOH wet mount, Gram's staining and 100µl saline aliquots for culture in solid media such as Sheep's Blood agar, Mac-Conkey agar, or Sabouraud's dextrose agar with chloramphenicol in a row of C-shaped streaks^{3,8,9}. In addition to above techniques modified AFB staining and parasitological study of wet mount from the saline aliquot. Sabouraud's Dextrose Agar plates were incubated at 28°C and were examined and observed at regular intervals for 1-2 wks for fungal culture.

Sheep's Blood agar, MacConkey agar Plates for bacterial culture were kept at 37°C and were observed for 01-02 days. Fungal growth was macroscopically identified on the basis of colony texture, color, diffusible pigments, growth rate, aerial and submerged hyphae and microscopically the morphological features like mycelium with lactophenol cotton blue, conidium, relationship between hyphae and fruiting bodies were noted from the culture. Slide cultures in potato dextrose agar and corn meal agar were used for observation of conidiogenesis of filamentous fungi for

identification. Identification of yeast-like fungi was done by Dalmau plate morphology, urease production, assimilation and fermentation of sugar. Fungal aetiology was considered when fungal growth was supported by positive direct microscopy or when growth of the same fungus was observed in more than two media or repeated isolation of the fungus on more than one occasion. Treatment was started with tropical antibacterial (Fluoroquinolones) and antifungal eye drop after the microbiological sample was taken and in most of the cases after getting the smear result. For fungal ulcers, 5% natamycin eye drop was started hourly. Periodical debridement was done unless corneal thinning precluded it till the ulcer bed was clean. All the cases which failed to improve within seven days were reviewed for other local or systemic causes. Cases with deep ulcers involving the posterior stroma, non-resolving hypopyon, hypopyon more than one-third of the anterior chamber, impending or actual corneal perforation or in cases where endophthalmitis was suspected, oral fluoroquinolone and/or oral fluconazole were given. Depending upon the fungal species and response to therapy natamycin eye drop was substituted with itraconazole (1%) or voriconazole (1%) eye drop and oral antifungal therapy.

Results:

In this present study corneal scrapings were collected from 300 numbers of patients with the clinical diagnosis of corneal ulcer with or without hypopyon those were presented in the department of Ophthalmology of Tripura Medical College, Hapania, and Tripura since last two year (1/11/2013 to 31/10/2015). A little hike of increase of incidence was observed during the month December to February. All of them were subjected for microbial investigation to find out the etiological agents. Of these 300 cases, 60% were found to be culture positive for bacteria and fungus and all the samples were positive for direct microscopy with gram's stain and KOH mount examination, 30% scrapings were culture sterile and also did not show any evidences of bacterial or fungal elements on direct microscopic examination for bacteria and fungus. Twelve cases among the 180 cases showed growth on culture though microscopically negative initially. The remaining ten cases did not show growth of any microbes despite of positive microscopic findings. All the samples might have received the treatment with topical antibiotics, antifungal, and corticosteroids time and again before presenting to hospital.

Table No.2: Age and sex distribution of corneal ulcers:

Age(yrs)	Male (%)	Female (%)	Total (%)	Bacteria (%)	Fungus (%)
0-12yrs	03(01)	03(01)	06(02)	03	00
13-24yrs	36 (12)	06(02)	42(14)	06	03
25-40yr	60 (20)	27(09)	87(29)	26	32
41-60yrs	54 (18)	42(14)	96(32)	44	26
61>yrs	51 (17)	18(06)	69(23)	23	17
Total	204(68%)	96(32%)	300	102 (56.6%)	78 (43.3%)

The Male patients were the most affected sex group accounts 68% in compare to female (32%). Total 180 cases were found to be culture positive with 102 bacterial and 78 fungal. The most common affected age group in male was 25-40 years where in female its 41-60 years. More than 75% patients of our study were from different villages of Tripura and majority were farmer, followed by house wife, daily worker (Labor) by profession.

Ocular trauma was found to be associated as major causes of corneal injury in 57.7% cases. Beside this, other predisposing factors like Diabetes, Exposure keratitis, Contact lens use, Corneal foreign body, dacryocystitis (NLD), dirty water, injury by nail, metallic substance, injury by flying insect etc. were noticed as being associated with cases with bacterial and fungal ulcers. The different predisposing factors and cause of ocular trauma associated with cases with bacterial and fungal ulcers are shown in Table 3.

Table. 3: The different predisposing factors associated with cases with bacterial and fungal ulcers:

Factors	Pure Bacterial corneal ulcers (n=91)	Pure Fungal corneal ulcer (n=70)	Mixed bacterial and fungal corneal ulcer (n=19)	Total (N=180)%
Topical antibiotic use	12	13	04	29(16.11%)
Ocular trauma	58	37	09	104(57.7%)
Diabetes	03	03	01	07(3.8%)
Exposure keratitis	03	02	00	05(2.7%)
Contact lens use	02	01	00	03(1.6%)
Corneal foreign body	04	05	02	11(6.1%)
Dacryocystitis(NLD)	07	08	03	18(10%)
*Others	02	01	0	03 (1.6%)

*Others include Post ocular surgery, Viral keratitis etc or without predisposing factor.

Cultures were positive in 180 cases of corneal ulcers. 102 (56.6%) patients had bacterial growth and 78(43.3%) fungal growth. In 19cases there was a growth of bacteria mixed with fungus. Detail growth bacterial and fungal isolates are shown in Table No.04. *Staphylococcus aureus* was the most commonly isolated bacterial organism 43 (23.8 %) of all positive bacterial cultures. The other isolated gram-positive organisms were *Staphylococcus epidermidis* 21 (11.67%), *Escherichiae coli* 14 (7.77%), *Pseudomonas spp* 11(6.11%), *Klebsiella spp* 9(5%)and *Acinetobater spp*(04) (2,22%) the most frequently occurring gram-negative organism was isolated from bacteriological growth cultures. (Table 4).

There were 78 fungal agents like *Aspergillus spp*-29(16.1%),*Candida albicans*-20(11.1) *Fusarium spp*-12(6.67%), *Curvularia*-06(3.33%), *Penicillium spp*-06(3.33%), *Rhodotorula spp*-03(1.67%) and *Trichosporon spp*-02(1.11%) isolated from 180 patients which also shown in Table 4. In mixed cases *Aspergillus spp*,*Candida albicans* and *Curvularia*- etc were. isolated along with bacteria.

Table. 4: Microbial agents isolated from corneal ulcers.

Name of bacteria isolated	No. of bacteria isolated, n=102(56.6%)	Name of fungus isolated	No. of fungus isolated, n=78 (43.3%)
<i>Staphylococcus aureus</i>	43(23.8)	<i>Aspergillus flavus</i> (10)	29(16.1)
		<i>Aspergillus fumigates</i> (11)	
		<i>Aspergillus niger</i> (08)	
<i>Staphylococcus epidermidis</i> (CNS)	21(11.67)	<i>Candida albicans</i>	20(11.11)

<i>Escherichiae coli</i>	14(7.77)	<i>Fusarium solani</i>	12(6.67)
<i>Pseudomonas spp</i>	11(6.11)	<i>Curvularia lunata</i>	06(3.33)
<i>Klebsiella spp</i>	09(5)	<i>Penicillium species</i>	06(3.33)
<i>Acinetobater spp</i>	04(2.22)	<i>Rhodotorula spp</i>	03(1.67)
		<i>Trichosporon spp</i>	02(1.11)
Total	102(56.6)		78(43.3)

Discussion:

Ocular fungal infections, or ophthalmic mycoses, are being increasingly recognized as an important cause of morbidity and Blindness. Corneal blindness is a major public health problem worldwide and infectious keratitis is one of the predominant causes incidence of which has increased worldwide in the last few years due to the improvement in microbiologic diagnostic techniques and because of introduction of new therapeutic measures such as widespread use of broad-spectrum antibiotics, immunosuppressive drugs and corticosteroids. In India, the prevalence of mycotic keratitis varies between 6-46.8 per cent in various regions¹⁰. In this study, Male (68%) age group is the most affective predominant group in comparison to female(32.%), this may be due to over exposure to environment, similar findings are present with most of the Indian study like Bharathi MJ et al¹¹, Jadhav S.V¹², Gopinathan U¹³ and many others. Chander J et al¹⁷ in his study, shown most frequently affected age group is between 21yr-50yrs which almost correlates with our findings, 41-60yrs age group is most affected group followed by 26-40 and 61>yrs age group in both sexes. But in contrast findings of Jadhav S.Vet al and Basak SK, does not match, their study shows 21-40yrs age group is affected group^{12, 15}

A significant increase of incidence of suppurative keratitis was observed during harvesting seasons of November-December than the wet, humid months of the year but few say during the hot and humid months^{14,15}. In this study we found the incidence increasing trend during the period from December-February as because this period coincided with the harvesting season in this region. In this present study we observed that many predisposing factors are responsible for ocular trauma of 57.7% which lead to corneal ulcer in majority of the cases. Vegetable matter mostly with leaf, paddy plants, paddy or paddy stalks responsible for causing supportive as well mycotic corneal ulcers in 40-50%cases alone. Besides Vegetable Stem, hay, Cow tail injury, Soil, muds, dirty water, Injury by nail, Metallic substance, Injury by flying insect etc. reported as frequent cause that responsible for formation of ulcer superficial to deep. Underlying disease like, Diabetes mellitus, Dacryocystitis or frequent injudicious use of tropical medication are underlying predisposing factors.. In our observation, villagers with their different profession like farmer, house wife, daily workers, agriculture workers etc are the commonest group of people are worst sufferer. Similar results are compared with the studies conducted like Madurai, Srinivasan M et al³, West Bengal, Basak SK et al¹⁵, Assam Reema Nath et al¹⁴

Cultures were positive in 180 cases of corneal ulcers where 102 (56.6%) patients had bacterial growth, 78(43.3%) had fungal growth and 19 cases had mixed bacterial and fungal growth. Our study results are almost correlated with the studies conducted in Madurai, Tamil Nadu 1997³, Ghana study in 1985¹⁹, Jadhav s.v study in Pune¹² Anil Kumar et al, in Gujarat, Western India¹⁹ etc. But it does not

approach the 80% or 81% isolation rate reported in Nepal 1991¹⁷ or Bangladesh 1994 study.¹⁸ Mono microbial infection was seen in majority (80.9%) of the cases the most common being bacterial (47.6%) followed by 33.3% fungal growth. Of these 180 cases of corneal ulcers 102 bacterial isolates were isolated from 102 bacterial growths. *Staphylococcus aureus* (23.8%) is being the most common bacterial agent isolated in our study followed by *Staphylococcus epidermidis*(11.67%), *Escherichiae coli* (7.77%), *Pseudomonas spp* (6.11%) and *Acinetobacter spp*(2.22%) respectively. Similar figures have been reported from Madurai(Srinivasan M et al)³, Andhra Pradesh(Kothapalli Sreelakshmi et al)¹⁶, in Gujarat(Anil Kumar et al)²⁰ and Ghana(Maria Hagan et al)¹⁹. As per study of Reema Nath et al¹⁴ and Basak SK et al¹⁵ mixed infection with bacteria is another problem in the diagnosis and treatment of fungal keratitis. Studies elsewhere from India reported mixed infection in 9.5-12% cases¹⁵. We too got few cases with mixed infection.

In our study there is a gradual trends of mycotic keratitis which is the major concern nowadays. *Aspergillus spp* (*Aspergillus flavus*, *fumigatus* & *niger*) were the major fungal etiological accounts 16.1% of the total 78 (43.3 %) fungal corneal ulcers followed by *Candida albicans*(11.1%), *Fusarium solani*(6.67%), *Penicillium species*(3.33%), *Rhodotorula spp*(1.67%)& *Trichosporon spp*(1.11%). Though our finding do not correlates the study Bharathi MJ et al¹¹ and Reema Nath et al¹⁴ with very high incidence (60.6%), but our results are within the Indian national range 6-46.8% which was shown in many Indian studies^{10,12,15,17,21}. Moreover this is our first experience, to see its gradual trends of mycotic keratitis in our centre which is already the major concern in world literature. 30% remained culture sterile, 07% culture negative despite strong clinical suspicion and positive microscopic result and 04% were culture positive but with negative microscopy in this present study. There may be five underlying causes (a)The reasons cultures to be sterile even when microscopy was positive could be that of patients were using tropical antibacterials, antifungals and corticosteroids before collection of corneal scraping. (b) Collection of inappropriate and inadequate quantity¹⁷. (c) Several limitations have been reported when such smears or mounts are used for corneal scrapes, including low sensitivity, frequent misinterpretation, presence of artifacts, and lack of detection of *Candida* and other yeasts²⁴. (d) KOH preparation tends to absorb carbon dioxide from air and form carbonate crystals thus reducing the effective hydroxide may lead to error in examination. (e) Moreover some medically important fungus produce metabolic end products which may results inhibition of growth²⁵. On direct microscopic examination with Gram's stain and 10% KOH mount examination techniques are still good, less expensive, users friendly and will remain gold standard test despite of its detection error.

Conclusion:

In summary, the suppurative keratitis is major public health concern. Bacteria may be the one of the major causes but this trends are reducing gradually because of peoples are mostly inhabiting rural areas, poverty, ignorance, involved in outdoor and agriculture activity as a result of which mycotic keratitis will continue to be an important cause for ocular morbidity. Present study provides regional trend of fungal etiology in fungal keratitis. In conclusion, clinical suspicion by the ophthalmologists should be confirmed by microbiological finding prior to initiating corticosteroids and anti-bacterial antibiotics. Precise identification of the causative fungus and institution of appropriate treatment strategy could save the blindness of the eye.

Reference(New article):

1. Geethakumari PV, Remya R, P S Girijadevi MS, Reena A MS, Bacterial Keratitis and Fungal Keratitis In South Kerala: A Comparative Study Kerala Journal of Ophthalmology, Vol. XXIII, No.1, Mar. 2011.
2. Whitcher JP, Srinivasan M, Upadhyay MP. Corneal blindness: a global perspective. Bull World Health Organ 2001; 79 : 214-21.
3. Srinivasan M, Gonzales CA, George C, Cevallos V, Mascarehas J M, Asokan B et al. Epidemiological and etiological diagnosis of corneal ulceration in Madurai, South India. Br J Ophthalmol. 1997;8:965-71.
4. Thylefors B, Negrel AD, Segaram PR, Dadzie KY. Available data on blindness (update 1994). Ophthalmic Epidemiology 1995; 2: 5-39. Garg P, Rao GN. Corneal Ulcer: Diagnosis and Management. Community Eye Health 1999; 12: 21-24.
5. Garg P, Rao GN. Corneal Ulcer: Diagnosis and Management. Community Eye Health 1999; 12: 21-24.
6. Thomas PA. Current perspective on ophthalmic mycosis. Clin Microbiol Rev 2003; 16: 730-97.
7. A. Saad-Hussein, H.M. El-Mofty and M.A. Hassanien :Climate change and predicted trend of fungal keratitis in Egypt, EMHJ • Vol. 17 No. 6 • 2011.
8. Sharma S, Athmanathan. Diagnostic procedures in infectious keratitis. In: Nema HV, Nema N, editors. Diagnostic Procedures in Ophthalmology . New Delhi: Jaypee Brothers Medical Publishers; 2002. pp 232-253.
9. Jones DB, Liesegang TJ, Robinson NM. Laboratory Diagnosis of Ocular Infections . Washington DC: Cumitech 13, American Society for Microbiology; 1981.
10. Rumpa Saha & Shukla Das, Mycological profile of infectious Keratitis from Delhi, Indian J Med Res 123, February 2006, pp 159-164.
11. Bharathi MJ, Ramakrishnan R, Vasu S, Meenakshi R, Palaniappan R. Aetiological diagnosis of microbial keratitis in South India: A study of 1618 cases. Indian J Med Microbiol 2002;20:19-24.
12. Jadhav S.V., gandham N.R., misra R.N., ujjagare M.T., sharma M. And sardar m. Prevalence Of Fungal Keratitis From Tertiary Care Hospital From Western Part Of India; International Journal of Microbiology Research; Volume 4, Issue 4, 2012, pp.-211-214.
13. Gopinathan U, Garg P, Fernandes M, Sharma S, Athmanathan S, Rao GN. The epidemiological features and laboratory results of fungal keratitis: A 10-year review at a referral eye care center in south India. Cornea 2002;21:555-59.
14. Reema Nath, Syamanta Baruah¹, Lahari Saikia, Bhanu Devi¹, A K Borthakur, J Mahanta²; Mycotic corneal ulcers in upper Assam; Indian Journal of Ophthalmology: September-October; Vol. 59 No. 5:367-71
15. Basak SK, Basak S, Mohanta A, Bhowmick A. Epidemiological and microbiological diagnosis of suppurative keratitis in Gangatic West Bengal, Eastern India. Indian J Ophthalmol 2005;53:17-22.
16. Kothapalli Sreelakshmi, Kandati Jithendra, Sarvepalli Skanda Harshita, Gujjula Chandra Sekhar , Buchineni Madhavulu, Pathapati Rama Mohan; A clinico-microbiological study of Suppurative Keratitis – one year experience in a tertiary care hospital of South India; International Journal of Sciences & Applied Research(IJSAR), 2(8), 2015; 23-30
17. Chander J, Sharma A. Prevalence of fungal corneal ulcers in North India. Infection 1994;22:207-9.
18. Upadhyay MP, Karmacharya PC, Koirala S, Tuladhar N, Bryan LE, Smolin G, et al. Epidemiologic characteristics, predisposing factors, and etiologic diagnosis of corneal ulceration in Nepal. Am J Ophthalmology 1991; 111: 92-99.
19. Dunlop AA, Wright ED, Howlader SA, Nazrul I, Husain R, McClellan K, Billson FA. Suppurative corneal ulceration in Bangladesh. A study of 142 cases examining the microbiological diagnosis, clinical and epidemiological features of bacterial and fungal keratitis; Aust N Z J Ophthalmol. 1994 May;22(2):105-10.
20. Maria Hagan, Elizabeth Wright, Mercy Newman, Paul Dolin, Gordon Johnson; Causes of suppurative keratitis in Ghana: British J Journal of Ophthalmology 1995; 79: 1024-1028.
21. Anil Kumar, Snehal Pandya, Ghanshyam Kavathia, Sejul Antala, Molly Madan, Tanuja Javdekar; Microbial keratitis in Gujarat, Western India: findings from 200 cases; Pan African Medical Journal. 2011; 10:48.
22. Talwar P, Sehgal SC. Mycotic infection of the eye in Chandigarh and neighbouring areas. Indian J Med Res 1978; 67 : 929-33.

23. B L Sherwal, AK Verma, Epidemiology of Ocular Infection Due to Bacteria and Fungus – A Prospective Study: JK Science: Vol. 10 No.3, July-September 2008.
24. Philip A. Thomas , Current Perspectives on Ophthalmic Mycoses Clinical Microbiology Reviews:Oct. 2003, p. 730–797
25. S Singh, PM Beena: Comparative study of different microscopic techniques and culture media for the isolation of dermatophytes; IJMM Year : 2003 ; Volume:21;Issue:1: 21-24.