



Clinical and Microbiological Profile of Infective Keratitis in Patients Presenting to a Tertiary Care Center in Jharkhand, India: A Cross-Sectional Study

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Abstract

Background and objective

Infective keratitis is a significant cause of ocular morbidity and visual impairment, particularly in developing countries. Early identification of the etiological agents and associated risk factors is essential for prompt and appropriate management. This study aimed to evaluate the clinical and microbiological profile of infective keratitis in patients presenting to a tertiary care center in Jharkhand and to assess the association between presenting visual acuity and the clinical etiological classification.

Methods

This hospital-based cross-sectional study was conducted from March to December 2025 at the Regional Institute of Ophthalmology, Rajendra Institute of Medical Sciences, Ranchi. A total of 168 patients with clinically diagnosed infective keratitis were included according to predefined inclusion and exclusion criteria. Detailed demographic and clinical histories were recorded, followed by a comprehensive ocular examination, including visual acuity assessment, slit-lamp biomicroscopy, and intraocular pressure measurement. Corneal scrapings were obtained under aseptic conditions and subjected to microbiological evaluation using Gram staining, a 10% potassium hydroxide (KOH) mount, and culture on appropriate media. Statistical analysis was performed using SPSS Statistics version 25.0 (IBM Corp., Armonk, NY), with a p-value of < 0.05 considered statistically significant.

Results

Among the 168 patients, males constituted the majority, accounting for 120 (71.43%) patients. The mean age of the study population was 49 ± 3.2 years. The most common risk factor was trauma, followed by diabetes mellitus. Most ulcers were located in the paracentral region, measured 2-5 mm in size, exhibited feathery margins, and demonstrated 20-50% stromal involvement. Fungal pathogens were the predominant isolates, with *Aspergillus flavus* identified in 38 (22.62%) patients, followed by *Fusarium* species in 30 (17.86%) patients. A statistically significant association was observed between a history of trauma and the occurrence of fungal keratitis.

Conclusions

Infective keratitis in this region was predominantly fungal in etiology, with *Aspergillus flavus* being the most commonly identified fungal pathogen. Early microbiological diagnosis and targeted antimicrobial therapy are essential for appropriate management and may help reduce visual morbidity.

Categories: Therapeutics, Ophthalmology, Infectious Disease

Keywords: aspergillus flavus, corneal scrapings, corneal ulcer, eastern india, fungal keratitis, fusarium, infective keratitis, microbial keratitis

Introduction

Infective keratitis, also known as microbial keratitis or corneal ulcer, is a vision-threatening ocular emergency that requires prompt diagnosis and management. It is characterized by a corneal epithelial defect with underlying stromal infiltration caused by pathogens, including bacteria, fungi, viruses, and protozoa.

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Given the cornea's vital role in maintaining transparency and refractive function, infection can lead to complications such as scarring, thinning, perforation, and irreversible visual impairment [1]. Globally, microbial keratitis remains a major cause of ocular morbidity and corneal blindness, particularly in developing countries, with an estimated 1.5-2 million new cases annually [2]. The burden is disproportionately higher in tropical and resource-limited settings due to favorable climatic conditions, poor socioeconomic status, and limited access to timely healthcare [3]. In India, corneal diseases are among the leading causes of monocular blindness [4], with infective keratitis contributing significantly to this burden [5].

The etiological profile varies geographically, with fungal keratitis predominating in tropical regions such as India, often associated with trauma from vegetative matter among agricultural workers [6]. Jharkhand, a predominantly rural and agrarian state, presents unique risk factors, including frequent occupational exposure to plant material, humid climatic conditions, delayed access to healthcare, and reliance on over-the-counter or traditional medications. Despite these factors, region-specific epidemiological data remain limited. Therefore, studying the clinical and microbiological profile of infective keratitis in Jharkhand is essential to identify local risk factors and microbial patterns, thereby enabling the development of region-specific treatment strategies, improving patient outcomes, and contributing to the prevention of corneal blindness [7,8]. The objective of this study was to assess the clinical and microbiological profiles of infective keratitis, focusing on bacterial and fungal pathogens and clinically diagnosed viral keratitis, among patients presenting to a tertiary care center in Jharkhand. In addition, this study aimed to assess the association between presenting visual acuity and clinical etiological classification.

Materials And Methods

This cross-sectional study was conducted at the Regional Institute of Ophthalmology, Rajendra Institute of Medical Sciences, Ranchi. Data were collected from 168 patients between March and December 2025. All cases of infective keratitis presenting to the outpatient and inpatient departments were included in the study. Patients with peripheral ulcerative keratitis, keratomalacia perforans, keratoconjunctivitis sicca, chemical injuries, vernal keratitis, immune-mediated keratitis, contact lens-associated keratitis, and those unwilling to participate were excluded. Ethical clearance was obtained from the Institutional Ethics Committee, and written informed consent was obtained from all participants.

Sample size calculation

The sample size was calculated using the formula for estimating a single population proportion:

$$n = \frac{Z^2 pq}{d^2}$$

Where n represents the required sample size, Z is the standard normal deviate corresponding to a 95% confidence level (1.96), p is the anticipated proportion, $q = 1 - p$, and d is the desired absolute precision. Based on a previously published study of microbial keratitis from Eastern India, in which fungal etiology accounted for 48.8% of cases, an anticipated proportion (p) of 48.8% was used [7]. Taking an absolute precision of 8%, the calculated minimum sample size was approximately 150 participants. Allowing for approximately 10% potential non-evaluable observations, the adjusted sample size was approximately 165 participants. A total of 168 patients with clinically diagnosed infective keratitis were enrolled. Microbiological findings were used to identify bacterial and fungal organisms, while viral keratitis was classified clinically.

Clinical assessment

A detailed history was recorded for each patient, including demographic information, age, sex, religion, address, occupation, socioeconomic status, history of present illness, past medical and surgical history, prior laboratory investigations, and any topical or systemic antimicrobial treatment received before presentation and corneal scraping. A comprehensive ocular examination was performed in all cases. Visual acuity was assessed using Snellen's chart, Landolt's C chart, or E chart, depending on patient cooperation. The eyelids were examined for abnormalities such as entropion, ectropion, trichiasis, and blepharitis, and the eyeball was evaluated for proptosis. Intraocular pressure was measured using applanation tonometry or digital palpation. Detailed anterior segment evaluation was performed using slit-lamp biomicroscopy. Corneal ulcers were stained with 2% fluorescein, and their size, site, shape, depth, margins, vascularization, infiltration, thinning or perforation, hypopyon characteristics, and presence of satellite lesions were documented.

Microbiological evaluation

Corneal scrapings were obtained under aseptic conditions from all 168 enrolled patients and subjected to microbiological evaluation. Smears were prepared for Gram staining and 10% potassium hydroxide (KOH) mount for preliminary detection of bacterial and fungal elements. Samples were inoculated onto blood agar, chocolate agar, nutrient agar, MacConkey agar, and Sabouraud dextrose agar. Bacterial isolates were further identified using coagulase testing and standard biochemical reactions where required. Fungal isolates were identified using conventional culture characteristics and microscopic morphology. Fungal growth on

Sabouraud dextrose agar was assessed based on colony morphology and microscopic characteristics, including the appearance and arrangement of fungal hyphae and reproductive structures, and isolates were identified to the genus/species level according to these conventional morphological characteristics.

No molecular or automated fungal identification method was used. Cases in which no organism was demonstrated on smear examination and no microbial growth was obtained on culture were classified as culture-negative/no microbial growth. These cases were distinguished from clinically diagnosed viral keratitis, which was assigned on the basis of characteristic clinical findings rather than by exclusion solely because of a negative culture. Smear findings and culture results were interpreted as distinct diagnostic assessments and were not considered interchangeable.

Clinical etiological classification

At presentation, cases were provisionally classified clinically as fungal, bacterial, or viral keratitis based on the overall clinical appearance of the corneal lesion, associated ocular findings, and relevant history. This clinical classification represented a presumptive diagnosis and was distinct from subsequent microbiological confirmation. Viral keratitis was diagnosed clinically based on the ocular examination findings documented at presentation. Clinically diagnosed viral keratitis was assigned when characteristic clinical findings supported a viral corneal etiology. Virological confirmation using polymerase chain reaction (PCR) or other virus-specific laboratory tests was not performed. Therefore, these cases were classified as clinically diagnosed viral keratitis rather than laboratory-confirmed viral infections. A negative bacterial/fungal microbiological result alone was not considered sufficient to classify a case as viral keratitis.

Cases without demonstrable organisms and without microbial growth that did not meet the clinical criteria for viral keratitis were categorized as culture-negative/no microbial growth. Thus, clinical and microbiological classifications were considered separate assessments. The final microbiological categories were mutually exclusive and comprised microbiologically confirmed fungal infection, microbiologically confirmed bacterial infection, clinically diagnosed viral keratitis, and no microbial growth. As the microbiological evaluation was based on conventional smear examination and culture methods for bacterial and fungal pathogens, viral keratitis was classified clinically; virus-specific laboratory testing was not performed, and no dedicated diagnostic methods for parasitic keratitis or fastidious bacterial pathogens were included in the study protocol. Therefore, these organisms were not systematically evaluated using pathogen-specific diagnostic techniques.

Statistical analysis

Data were analyzed using SPSS Statistics v25.0 (IBM Corp., Armonk, NY). Categorical variables were expressed as frequencies and percentages. The chi-square test was used for categorical data, and a p -value < 0.05 was considered statistically significant. The association between history of ocular trauma (present or absent) and clinical etiological classification (fungal, bacterial, or viral keratitis) was assessed using the Pearson chi-square test of independence. A two-sided p -value < 0.05 was considered statistically significant.

Results

A total of 168 patients with infective keratitis were included in the study. The patients' ages ranged from 10 to 87 years, with a mean of 49 ± 3.2 years. The largest proportion of patients was aged > 50 years (78; 46.43%), followed by those aged 41–50 years (44; 26.19%), 31–40 years (29; 17.26%), 21–30 years (13; 7.74%), and 10–20 years (4; 2.38%). The demographic characteristics and laterality of the study population are presented in Table 1. Males constituted the majority of the study population (120; 71.43%), while 48 (28.57%) were females, giving a male-to-female ratio of approximately 2.5:1. Most patients were from rural areas (132; 78.57%), whereas 36 (21.43%) were from urban areas. Among the reported occupational groups, farmers constituted the largest group (81; 48.2%), followed by laborers (30; 17.9%), mechanics (15; 8.9%), and housewives (15; 8.9%). All cases had unilateral involvement: the right eye in 80 patients (47.62%) and the left eye in 88 (52.38%); no bilateral cases were observed. Based on clinical assessment at presentation, 102 (60.71%) patients were provisionally classified as having fungal keratitis, 26 (15.48%) as bacterial keratitis, and 40 (23.81%) as viral keratitis. Before corneal scraping, 60 (35.7%) patients had received topical and/or systemic antimicrobial therapy, whereas 108 (64.3%) patients had not received antimicrobial treatment before sampling. Prior antimicrobial exposure may have reduced microbiological yield and may partly explain the discrepancy between clinical classification and microbiological confirmation.

Characteristic	N (%)
Age group, years	
10–20	4 (2.38)
21–30	13 (7.74)
31–40	29 (17.26)
41–50	44 (26.19)
> 50	78 (46.43)
Sex	
Male	120 (71.43)
Female	48 (28.57)
Residence	
Rural	132 (78.57)
Urban	36 (21.43)
Laterality	
Right eye	80 (47.62)
Left eye	88 (52.38)
Bilateral	0 (0.00)

TABLE 1: Demographic characteristics of the study population

Percentages are calculated using the total study population (N = 168)

The findings summarized in Table 2 depict that microbiological evaluation identified fungal organisms in 78 (46.43%) patients, including *Aspergillus flavus* (38; 22.62%), *Fusarium* species (30; 17.86%), *Aspergillus niger* (5; 2.98%), *Bipolaris spicifera* (4; 2.38%), and *Candida albicans* (1; 0.60%). Of the 168 patients enrolled with clinically diagnosed infective keratitis, bacterial organisms were identified in 21 (12.50%) patients and fungal organisms in 78 (46.43%), giving a total of 99 (58.93%) microbiologically positive cases. No microbial growth was observed in 29 (17.26%) patients. The remaining 40 (23.81%) patients were classified as having clinically diagnosed viral keratitis without virological confirmation. Culture-negative cases were distinguished from clinically diagnosed viral cases, and the absence of microbial growth was not interpreted as definitive evidence of a noninfectious etiology. Therefore, microbiological confirmation was available for 99 patients, whereas the etiological classification of the remaining patients was based on clinical assessment or the absence of microbial growth.

Microbiological category	Organism/diagnosis	N (%)
Fungal	<i>Aspergillus flavus</i>	38 (22.62)
	<i>Fusarium</i> species	30 (17.86)
	<i>Aspergillus niger</i>	5 (2.98)
	<i>Bipolaris spicifera</i>	4 (2.38)
	<i>Candida albicans</i>	1 (0.60)
Total fungal		78 (46.43)
Bacterial	<i>Staphylococcus aureus</i>	14 (8.33)
	<i>Staphylococcus epidermidis</i>	6 (3.57)
	<i>Pseudomonas</i> species	1 (0.60)
Total bacterial		21 (12.50)
Viral	Clinically diagnosed viral keratitis	40 (23.81)
No microbial growth	No growth on culture	29 (17.26)
Total		168 (100.00)

TABLE 2: Microbiological findings and clinical classification of patients with infective keratitis

Percentages are calculated using the total study population (N = 168). Of the 168 patients, 99 had bacterial or fungal organisms identified, 29 had no microbial growth, and 40 were clinically diagnosed with viral keratitis without virological confirmation

The distribution of presenting visual acuity according to clinical etiological category is shown in Table 3. Visual acuity categories were classified as mild (6/9-6/18), moderate (6/18-6/60), severe (6/60-3/60), and blindness (worse than 3/60 to no light perception). The etiological categories in this table represent the presumptive clinical classification at presentation and should not be interpreted as microbiological confirmation. Mild visual impairment was observed in nine patients (5.4%), moderate in 48 (28.6%), severe in 60 (35.7%), and blindness in 51 (30.4%). Clinically diagnosed viral keratitis was the predominant clinical etiological category among patients with mild and moderate visual impairment, whereas clinically classified fungal keratitis predominated among those with severe visual impairment and blindness.

Presenting visual acuity	Fungal keratitis, n (%)	Bacterial keratitis, n (%)	Viral keratitis, n (%)	Total, n (%)
Mild	1 (11.1)	0 (0.0)	8 (88.9)	9 (100.0)
Moderate	19 (39.6)	4 (8.3)	25 (52.1)	48 (100.0)
Severe	42 (70.0)	12 (20.0)	6 (10.0)	60 (100.0)
Blindness	40 (78.4)	10 (19.6)	1 (2.0)	51 (100.0)
Total	102 (60.71)	26 (15.48)	40 (23.81)	168 (100.0)

TABLE 3: Presenting visual acuity according to clinical etiological classification

Percentages within each visual-acuity category are row percentages, while percentages in the total row are calculated using the overall study population (N = 168). The association between the presenting visual acuity category and the clinical etiological category was assessed using the Pearson chi-square test ($\chi^2 = 62.15$, df = 6, $p < 0.001$). A two-sided $p < 0.05$ was considered statistically significant

A statistically significant association was observed between history of ocular trauma and clinical etiological classification (Pearson chi-square test: $\chi^2 = 30.60$, df = 2, $p < 0.001$) as shown in Table 4. A history of ocular trauma was reported more frequently among patients clinically classified as having fungal keratitis than among those classified as having bacterial or viral keratitis.

History of trauma	Fungal keratitis (n = 102)	Bacterial keratitis (n = 26)	Viral keratitis (n = 40)	Total (N = 168)
Present	72 (70.6%)	12 (46.2%)	8 (20.0%)	92 (54.8%)
Absent	30 (29.4%)	14 (53.8%)	32 (80.0%)	76 (45.2%)
Total	102 (100%)	26 (100%)	40 (100%)	168 (100%)

TABLE 4: Association between trauma history and clinical etiological classification

Percentages in the etiological columns are calculated within each clinical category. Pearson chi-square test of independence was employed. Chi-square statistic: $\chi^2 = 30.60$, $df = 2$, $p < 0.001$

As is clear from Table 5, fungal keratitis was the predominant clinical etiological classification, accounting for 102 (60.71%) patients. However, fungal organisms were microbiologically identified in 78 (46.43%) patients, highlighting the distinction between clinical classification and microbiological confirmation. Thus, the clinical classification and microbiological findings were not identical, with 24 clinically classified fungal cases and five clinically classified bacterial cases lacking corresponding microbiological confirmation. The microbiological classification was mutually exclusive: cases were categorized as microbiologically confirmed fungal keratitis, microbiologically confirmed bacterial keratitis, clinically diagnosed viral keratitis, or no microbial growth. Bacterial and fungal organisms were identified from corneal specimens by smear examination and/or culture. Viral keratitis was diagnosed clinically based on characteristic clinical findings. Cases with no demonstrable organism and no microbial growth were categorized as having no microbial growth and were not considered microbiologically confirmed bacterial or fungal keratitis. No statistical test was applied to the descriptive microbiological distribution.

Classification	Patients	Percentage
Microbiologically identified fungal organisms	78	46.43%
Microbiologically identified bacterial organisms	21	12.50%
Total microbiologically positive	99	58.93%
No microbial growth	29	17.26%
Clinically diagnosed viral keratitis	40	23.81%
Total enrolled	168	100%

TABLE 5: Overall distribution of the cases

Discussion

Infective keratitis remains a major cause of corneal morbidity and preventable monocular blindness in tropical developing regions, with a disproportionately higher burden among rural, agriculturally dependent populations. The present study from Jharkhand highlights the influence of environmental exposure, occupational risk, and limited access to specialized ophthalmic care on disease patterns. The region’s tropical climate, characterized by high temperatures, substantial rainfall, and predominant agrarian activity, creates favorable conditions for fungal keratitis, particularly following vegetative ocular trauma [9,10,11]. Poor awareness regarding early symptoms, reliance on traditional remedies, and delayed referral to tertiary centers may have contributed to the advanced disease severity observed at presentation [5,12]; however, these factors were not directly evaluated in this cross-sectional study.

Fungal keratitis was the predominant clinical etiological classification, accounting for 102 (60.7%) patients, consistent with findings from eastern India and other tropical regions [13,14]. However, fungal organisms were microbiologically demonstrated in 78 (46.4%) patients, highlighting the distinction between clinical suspicion and laboratory confirmation. Among the microbiologically identified fungal organisms, *Aspergillus* and *Fusarium* species predominated. Similar microbiological profiles have been reported in West Bengal and Bihar [15,16]. In contrast, studies from other regions of India, such as north Karnataka, and neighboring countries like Nepal demonstrate a higher prevalence of bacterial keratitis, highlighting significant geographical variation [17,18].

This regional variability underscores the importance of tailoring empirical therapy based on local

microbiological patterns rather than adopting a uniform treatment approach. The disease predominantly affected males, elderly individuals, and rural populations, with farmers constituting a major proportion, reflecting occupational exposure risks, particularly trauma with vegetative matter. Most patients presented with advanced disease and severe visual impairment, which may be related to delayed healthcare access and prior treatment practices, including prior inappropriate use of topical medications such as corticosteroids [19,20], although these factors were not directly evaluated in the present study.

Microbiologically, filamentous fungi were the most common isolates, while Gram-positive cocci predominated among bacterial pathogens. Smear examination remains a valuable rapid diagnostic tool in resource-limited settings, facilitating early targeted therapy [21]. Prior antimicrobial exposure may have influenced the microbiological findings. In our study, 35.7% of patients had received antimicrobial therapy before corneal scraping, which may have reduced culture yield and partly contributed to the lower proportion of microbiologically confirmed bacterial and fungal infections compared with the corresponding clinical classifications.

Limitations

As this was a hospital-based study, the possibility of referral bias could not be excluded, and the findings may not fully reflect the incidence or disease pattern in the general community. Seasonal variations were not assessed in detail because of the relatively short study duration. In addition, molecular and advanced fungal identification facilities were unavailable; therefore, fungal identification was based on conventional smear and culture-based morphological methods, which may have limited the identification of fastidious, uncommon, or atypical organisms. Prior antimicrobial exposure may also have influenced culture yield and resulted in underestimation of microbiologically confirmed infections.

Although Gram staining and 10% KOH mount were performed as part of the microbiological evaluation, the corresponding smear findings were not available for separate quantitative reporting. Therefore, the contribution of direct smear microscopy to etiological identification could not be independently assessed, and the microbiological interpretation was based on the available culture findings and clinical classification. The absence of virus-specific testing and dedicated diagnostic methods for parasitic keratitis and fastidious bacterial pathogens may have limited the identification of these etiologies. Consequently, some cases may have remained etiologically unconfirmed, and the findings should be interpreted in the context of the diagnostic methods employed.

Conclusions

Fungal keratitis was the most common clinical etiological classification in this study. Among patients with clinically diagnosed infective keratitis, fungal organisms were the most frequently identified microorganisms on microbiological evaluation, with filamentous fungi, particularly *Aspergillus* and *Fusarium* species, being the most frequently identified pathogens. However, viral keratitis was diagnosed clinically without laboratory confirmation, and no microbial growth was observed in a subset of patients. These findings should be interpreted in light of the distinction between microbiologically identified organisms and clinical etiological classification. Males and individuals engaged in manual labor constituted the majority of affected patients, and a history of corneal trauma, particularly trauma involving vegetative matter, was an important predisposing factor. Most patients presented with moderate to severe visual impairment and substantial stromal involvement. The observed association between a history of ocular trauma and clinical fungal keratitis underscores the importance of documenting trauma history during clinical evaluation. However, the cross-sectional design does not establish a causal relationship. Microbiological assessment was essential for establishing the etiological diagnosis and guiding appropriate treatment in these patients.

Additional Information

Author Contributions

All authors have reviewed the final version to be published and agreed to be accountable for all aspects of the work.

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Disclosures

Human subjects: Informed consent for treatment and open access publication was obtained or waived by all participants in this study. Institutional Ethics Committee RIMS, Ranchi issued approval 157. **Animal subjects:** All authors have confirmed that this study did not involve animal subjects or tissue. **Conflicts of interest:** In compliance with the ICMJE uniform disclosure form, all authors declare the following: **Payment/services info:** All authors have declared that no financial support was received from any organization for the submitted work. **Financial relationships:** All authors have declared that they have no financial relationships at present or within the previous three years with any organizations that might have an interest in the submitted work. **Other relationships:** All authors have declared that there are no other relationships or activities that could appear to have influenced the submitted work.

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