A PROSPECTIVE STUDY OF EPIDEMIOLOGICAL FEATURES, MICROBIOLOGICAL DIAGNOSIS & TREATMENT OUTCOME OF INFECTIVE KERATITIS

Ophthalmology

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ABSTRACT

Aim: To study the epidemiological characteristics, microbiological profile, and treatment outcome of patients with suspected microbial keratitis in the region of Udaipur, Rajasthan.

Materials & methods: A prospective, analytical, hospital-based study conducted, all the patients presenting with corneal stromal infiltrate were enrolled for study and underwent standard microbiologic evaluation of their corneal scrapings, and smear and culture-guided antimicrobial therapy. Follow up was done for 1 month and outcome were evaluated.

Results: In our study, 60 patients with infective keratitis were enrolled. 38 (63.3%) were male patients while 22 (36.7%) patients were females. 58% people with infective keratitis had trauma as a leading predisposing factor. 19 patients were diagnosed viral keratitis and 41 (68.33%) patients were found to be culture positive. Among the 41 culture positive patients 31.7% were bacterial, 30% were fungal, 6.7% were mixed. The majority of our patients 44 (73.3%) healed with the corneal scar, 12 [20%] healed with clear cornea.

Conclusion: Our study observed trauma with vegetative material is the most common predisposing for infective keratitis. Males were affected more than females. Among organisms coagulase negative staphylococci and aspergillus were most common bacterial & fungal isolates respectively. Herpes simplex virus was the most common viral isolate. Most of the patients of infective keratitis healed with the corneal scar.

KEYWORDS

Infective Keratitis, Microbial Diagnosis, Treatment Outcome

INTRODUCTION

Keratitis is the term applied for inflammations of the cornea.⁽¹⁾ Keratitis could be infective and non-infective. Differentiating between them is crucial in managing both. Careful clinical examination, aided by laboratory investigations, could help in correct diagnosis and proper management.

Corneal blindness ranks next to cataract among the major causes of blindness. Ocular trauma, as well as corneal ulceration, is responsible for most of these cases of corneal blindness. In the developing world, infectious corneal ulcer is a leading cause of prolonged ocular morbidity and visual loss.⁽¹⁾ Corneal blindness is responsible for 1.5-2 million new cases of monocular blindness every year.⁽²⁾ In India, approximately 6.8 million people are suffering from corneal blindness. There is a significant variation in the prevalence of corneal ulceration in different parts of the world. This can be attributed to difference in climatic conditions, difference in occupation as well as other socioeconomic factors.^(4,5) Population-based studies conducted found that the incidence of corneal ulceration is 10 times higher in India as compared to the Western countries.^(6,7)

In this study, we aim to understand the epidemiology, the predisposing factors, microbiological profile and the outcome of management of infective corneal ulcers presenting to our hospital, which is a tertiary care hospital located in Udaipur, Rajasthan.

MATERIAL AND METHODS

In this prospective hospital based study, a total of 60 clinically diagnosed cases of infective keratitis, who attended the Ophthalmology Out-Patients Department of R.N.T. Medical College, Udaipur, India were studied over a period of fifteen months, after obtaining informed patient consent and approval by Institutional Ethical Committee.

Patient Selection:

All patients with infectious corneal ulcers and suspected viral keratitis presenting to the ophthalmology outdoor at MBGH, Udaipur from September 2014 to December 2015 were included in the study.

Corneal ulceration was defined as a loss in the continuity of corneal epithelium with underlying stromal necrosis along with associated signs of inflammation, with or without hypopyon.

A detailed history was obtained from each patient, with special emphasis on the patient's occupation and any prior history of trauma. If present, the mode of injury, as well the details of previous treatment taken, was noted. Special inquiry was made regarding the use of steroid eye drops. History of any antecedent febrile illness as well as the history of contact lens wear was also sought. History of any associated systemic illness such as diabetes was taken. The nature and duration of complaints was noted. The best-corrected visual acuity (BCVA) was recorded. Each patient was subjected to a detailed slit-lamp bio-microscopic examination, with special emphasis on the cornea. The details of the corneal ulcer - including the location, size, shape, depth of the ulcer were noted after staining with fluorescein, nature of infiltrate, margins of the ulcer, presence of any satellite lesions, corneal vascularization, and hypopyon were noted. Photographic documentation of the corneal ulcer on slit-lamp imaging system was done at the time of initial presentation, as well as on each follow-up visit. Associated ocular conditions such as blepharitis, conjunctivitis, dacryocystitis, dry eyes, corneal sensations, or ocular leprosy were noted.

For viral keratitis laboratory tests are not an absolute requisite for the diagnosis of HSV infection as clinical features are often highly characteristic. Various clinical findings suggestive of epithelial, stromal and endothelial infiltration were noted. Specific fluorescein staining pattern of dendritic and geographical ulcer were drawn.

Corneal scraping was performed with full aseptic precautions, after anesthetizing the cornea with 2% proparacaine without preservative, with a sterile No. 15 Bard-Parker blade. The material was obtained by gently scraping the leading edge and base of the ulcer. The material was smeared on two slides - One for Gram stain and other as 10% potassium hydroxide (KOH) wet mount.^{9,10} For culture and sensitivity, the material was also directly inoculated by multiple C-shaped streaks, on glucose broth, nutrient agar and Sabouraud dextrose agar (SDA). The laboratory diagnosis was performed using standard protocols.

All bacterial cultures were inoculated aerobically at 37°C. Cultures on blood agar, nutrient agar and McKonkey agar were evaluated at 24 hours and at 48 hours and then discarded if there was no growth. Fungal cultures inoculated onto SDA were incubated at 27°C, examined daily, and discarded at 3 weeks if no growth was present.

Specific identification of bacterial pathogen was based on microscopic morphology, staining characteristics, colony growth characteristics and biochemical properties using standard laboratory criteria. Fungi were identified by their colony characteristics on SDA and by the morphological appearance of the spores in lactophenol cotton blue stain.

Treatment was started empirically on the bases of clinical presentation after taking the corneal scraping, on the same day of presentation.

Follow-up of the patient was documented at day 4, 1 week, 2 weeks and 1 month, respectively, though the actual follow-up was done more frequently.

On follow-up, we recorded the patient's BCVA, corneal ulcer size, hypopyon (present/absent/decreased), infiltration (increased/decreased) and symptomatic relief as reported by the patient. The patient was said to have improved if the size of the ulcer, hypopyon, infiltration had decreased and patient's symptoms had improved.

RESULTS

The study was conducted in RNT Medical College, Udaipur during September 2014 to December 2015. Out of 60 patients 38 (63.3%) were males and 22 (36.7%) were female. The overall male to female ratio was 1.72:1. Maximum no. of patients were in the age group 21 to 40 years [43.3%] followed by 41 to 60 years (36.7%) (figure.1).

age d stribution

Occupation of a person also has a bearing on the causation of corneal ulcer in many cases. In our study, we found that 18 (30%) out of 60 patients were involved in farming activities. 14 (23.3%) patients were involved in labour. Thus, incidence of keratitis is higher among the outdoor workers than indoor or desk job. (Table 2)

Table No.2

Occupation	Frequency	Percent
Farmer	18	30.0
Labour	14	23.3
Businessman and office worker	6	10.0
House wife	8	13.3
Student	8	13.3
Unemployed	6	10.0
Total	60	100.0

The most common predisposing factor observed in the study responsible for keratitis was trauma which was seen in 58.3% of patients. Other conditions predisposing to keratitis were diabetes (3.3%), dry eye (1.7%), dacryo-cystitis (1.7%) and conjunctivitis (1.7%).

Among the cases of ocular trauma, history of vegetative injury was the leading cause seen in as many as 13 (21.7%) patients. A history of injury by other wooden material was seen in 5 (8.3%) patients. History of injury by unknown foreign body was in 9 (15%). Injury by materials such as stone/sand particles was seen in 1 (1.7%) patients. [Graph 1]

Graph 1

In our study Out of the 60 patients 19 [31.7%] were diagnosed as viral keratitis clinically and 41 (68.3%) patients were found to be culture positive. Among the 41 culture positive patients, 19 (31.7%) patients were positive for bacteria, while 18 (30.0%) patients were positive for fungi and 4 (6.6%) were mixed.

Graph 2

Bacterial microorganism were cultured from 23 patients of corneal ulcer out of which 19 (82.6%) were pure isolates and remaining 4 (17.4%) were cultured with fungi. Most commonly isolated gram +ve organism was coagulase -ve staph 11/23 (47.83%) followed by staph aureus (34.8%). Among cultured fungal isolates aspergillus [12/22-54.5%] is most common followed by Fusarium [6/22-27.3%].

Fig.1. Bacterial Keratitis

Fig.2.fungal Keratitis

Table No.4

Bacterial micro-organism	Pure isolate	Mix with fungi	Total	P value
Gram +ve cocci	Staph aureus	7	1	0.003
		30.43%	4.34%	
	Coagulase -ve staph	8	3	
		34.8%	13.04%	
			47.83%	
Gram -ve bacilli	Pseudomonas	2	0	0.003
		8.69%	0%	
			8.69%	
Total	Micrococci	1	0	0.003
		4.34%	0%	
Total		4.34%	0%	0.003
			4.34%	
Total		19	4	
		82.60%	17.4%	100.0%

Table No.5

Fungal micro-organism		Pure Fungal	Mixed	Total
Filamentous hyphae	Aspergillus	10	2	12
		45.4%	9.1%	54.5%
	Fusarium	6	0	6
		27.3%	0%	27.3%
	Penicillium	0	1	1
		0.0%	4.54%	4.54%
Unidentified	Unidentified	2	1	3
		9.1%	4.54%	13.6%
Total		18	4	22
		81.8%	18.2%	100%

Fig.3. Pigmented Fungal Ulcer

Among 19 clinically diagnosed viral keratitis 16 [84.2%] were HSV and remaining 3[15.8%] were HZO.

Fig.4. Viral Dendritic Ulcer

Fig.5.viral Superficial Punctate Keratitis.

All patients were started on medical therapy initially and outcome was noted on 28th day. 44 [73.3%] patients healed with the corneal scar, 12 [20%] healed with clear cornea; 3[5%] were not responding to medical treatment with unknown reason.

Most of the viral keratitis (12/19)(63.15%) healed with the clear cornea but almost all ulcerative keratitis of bacterial and fungal origin healed with the scar. One mixed isolate patient required surgical intervention in the form of penetrating keratoplasty. **Antibiotic sensitivity pattern of Gram Positive cocci and Gram Negative bacilli are shown in table no.6**

Table No.6

	Amoxicillin	Oxacillin	Cefazoline	Vancomycin	Ciprofloxacin	Moxifloxacin	Amikacin	Gentamycin	Azithro
Staph aureus (8)	5	8	8	8	5	8	8	2	5
Coagulase -ve staph (11)	7	9	11	11	7	11	11	8	8
Pneumococci (2)	2	2	2	2	1	2	2	0	2
Micrococci(1)	1	1	1	1	1	1	1	1	1
Pseudomonas (1)	0	0	1	1	0	1	1	0	1

DISCUSSION

In this study, we analysed epidemiological features, laboratory diagnosis and ultimate treatment outcome of the infective keratitis patients visited in our OPD at RNT Medical College, Udaipur.

In the present study infective keratitis was seen in all age groups with preponderance among physically active adults; higher in males [63%] than in females [37%] as observed in study of Kashmir [65%]^[10], south India [56.6%]^[8]; and Ghana [69.3%].^[5]

In our study trauma was the most common predisposing factor [58%] matching with other studies from study of Madurai [65.4%]^[12] and more than in Ghana [39%]^[5]. Corneal injury with vegetative matter was the most common followed by unknown foreign body and wooden objects as found in the study of Basak et al^[3].

There was higher incidence of keratitis among farmer [30%] followed by labourer.

[23.3%], housewives [13.3%] and students [13.3%] in this This implies significant association between occupation and infectious keratitis.

Microorganisms were isolated from 41 corneas that were cultured. These compares favourably with the study in Ghana [58%]^[5], Madurai [68.4%]^[12], west Bengal [67.7%].^[3]

In our study bacterial keratitis were positive in 19/60 [31.7%] cases but among the 41 patients of ulcerative keratitis culture proven bacterial keratitis found in 19/41 [46.34%] which compares well with other studies across India like study done in Kashmir [40%]^[10], Madurai[32.3%]^[12], Bharathi MJ, Ramakrishnan R et al [32.7%]^[8]. Bacteria are responsible for a larger proportion of corneal ulceration in temperate climates than in tropical regions such as South India. The incidence of Gram-positive cocci (97%) of bacterial etiology, 22/41 [53.65%] of total corneal ulcers) coincides with studies of Basak et al. [71%]^[3] and Shrinivasan et al. [79%]^[12], predominance of coagulase negative staphylococci [47.8% of total gram +ve cocci isolates] coincides well with the study done in Ghana [43%]^[5].

Gram-positive bacilli were not observed in the present study. Only one isolate of gram -ve bacillus pseudomonas was found in our study[5.8%] which was as low as in study done in Kashmir[12%]^[10]. From the antibiotic sensitivity pattern, it appears that approximately all the patients with bacterial etiology would have responded to eyedrop Moxifloxacin monotherapy and if fortified cefazoline, fortified vancomycin and fortified amikacin antibiotic eyedrops therapy were employed. We feel that the number of samples studied is relatively small to make a definite recommendation. However, Moxifloxacin may be used as a monotherapy in peripheral or small ulcers and; fortified cefazoline or fortified vancomycin and fortified amikacin in the central or large ulcers may be used as first line therapy in the Udaipur region.

Fungal growth seen in 20% of total keratitis patients, 29.2% in the corneal ulcer patients. Almost half (43.9%) of all corneal ulcers with positive cultures were fungal in origin. If the 4 mixed infections (9.75%) are also considered to be primarily fungal for treatment purposes and they are added to the pure fungal cases, 53.65% of all culture positive corneal ulcers grew fungal pathogens. This figure approaches the fungal isolation rate in study at Ghana^[5] where cultured fungi from 56% of 114 patients who had positive cultures and study at Madurai [57%]^[12] and west Bengal [62.7%]^[3] but differ from

study of Barati MJ at south India [32%]^[8], [34.8%] at Ahmedabad^[7] and Bharati MJ et al (2003) in south India [34.4%]^[8] which all show lower incidence and very low in Nepal [17.6%]^[5]. *Aspergillus* species (54.5%) of total fungal etiology in the present study matches other studies at west Bengal [59.8%]^[5] and Kashmir [40%]^[10]. Incidence is markedly low in Madurai [16.1%]^[12] and south India [24.3% and 26%]^[2,11]. *Fusarium* incidence of 28.6% was nearly similar to that was in other studies done in West Bengal [21.2%]^[3] and Ahmedabad [22.5%]^[7]. However, it was predominant in South India and Ghana^[4,5,11,12] in the range of 40 to 50 %. Most of these fungal growths were seen in farmers because of injury with vegetative matter. Yeast infection was not found in this study as supported by Bharati et al^[2] and Shrinivasan et al^[12].

Among the total 60 keratitis patients 19 were clinically diagnosed as viral keratitis. Any lab investigation was not done for the diagnosis of viral keratitis. Among 19 clinically diagnosed viral keratitis 16 [84.2%] were showing the signs of herpes simplex epithelial and stromal viral keratitis. Remaining 3 [15.8%] were presented with clinical signs of herpes zoster ophthalmicus associated with facial herpes zoster. 89.4% patients of viral keratitis did not show any presence of trauma. This higher incidence of viral keratitis [31.66%] may be due higher incidence of immunocompromised status. Almost all the patients of viral keratitis respond well to the medical treatment. As in our study all the patients of keratitis started with the medical treatment initially showing results that all the patient of ulcerative keratitis (100%) healed with the corneal scar compared to viral keratitis [12/19 (63.2%)] healed with the clear cornea and [7/19 (36.8%)] healed with corneal scar. Three patients one is fungal, one mixed and one is non-specific where not responding to medical treatment and one case of mixed [bacterial + fungal] taken for surgical treatment [penetrating keratoplasty]. Most of the fungal keratitis patients taken longer duration to heal than bacterial keratitis and most fungal keratitis were unresponsive to treatment. This study shows that although bacterial keratitis can be treated effectively, the treatment of fungal keratitis remains a challenge supported by study done by Usha Gopinathan et al^[9].

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