Original research article

Clinical and microbiological profile of infectious corneal ulcers and its seasonal trend in a tertiary care hospital of south west Bihar.

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Received: 14-08-2020 / Revised: 10-09-2020 / Accepted: 26-10-2020

Abstract

Aim; To identify the microbiological profile and associated risk factors for microbial corneal ulcer in a tertiary care hospital of South West Bihar. **Methods**; In this prospective study conducted at Narayan Medical College and Hospital, Sasaram, Bihar, India. Total 61 patients with a clinical diagnosis of corneal ulcer were included. After detail history and slit lamp evaluation of corneal ulcer, corneal scraping was performed and subjected to microbiological analysis including Gram stain, potassium hydroxide(KOH) wet mount and bacterial and fungal culture. **Results;** Among total 61 patients included in our study, male to female ratio is almost equal and the most common affected age group was 41-60 years. Commonest profession affected was agriculture workers in 77% of cases and the commonest risk factor associated was trauma while working in agriculture fields in 44% of cases. The peak in the incidence of corneal ulcer cases were during harvesting of paddy crops in winter months. Forty eight percent of the ulcers were found to be culture positive. Among culture positive patients 79% were fungal and rest 21% were bacterial. Fusarium spp was the most common fungal isolate(48%) followed by Aspergillus spp (26%).Among bacteria S. aureus was the most common organism(66.6%) isolated. **Conclusion**; :- In our study, we found that that in region of south west Bihar , fungal (specially Fusarium spp) corneal ulcers predominate, as the majority of patients present with a history of antecedent vegetative trauma mainly during harvesting of paddy crops in winter season. Managing infective suppurative corneal ulcers with timely and appropriate antimicrobial therapy will help to prevent the cases of corneal ulcer and it's sequel.

Key words: Corneal ulcer, Microbiological profile, Fusarium, Seasonal trend

Introduction

[1] The prevalence of individual pathogens largely depends on geographical and climatic factors.

Corneal ulcer occurs mainly in the warm climates and coincides with seasonal increase in temperature and humidity.

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Corneal ulcer, a major cause of monocular blindness and visual disability in developing countries.

Trauma, particularly by vegetative or soil matter, seems to be the most common predisposing factor for corneal ulcer [2]. Apart from that, injudicious use of topical corticosteroids and antibacterial agents for external ocular disease enhances the risk further. [3] Several investigators have reported the prevalence of bacterial and fungal pathogens isolated from ulcerated corneas. Corneal ulcer is a major health problem in developing world causing prolonged

ocular morbidity and loss of vision. [4] The major morbidity from infectious keratitis is due to corneal ulceration and subsequent perforation which can lead to endophthalmitis, or visual loss from severe scarring and vascularization.[5]. Due to large agrarian population and environmental factors, corneal ulcer is common in India. The males are more commonly affected as compared to females due to their higher outdoor exposure. In northern India Aspergillus is commonest cause where as in south India, it is Fusarium, which is the most common. Corneal ulcer is an ophthalmic condition requiring prompt medical attention. Thus precise knowledge of the causative agents and their susceptibility patterns is important for deciding the proper course of treatment [6]. Corneal ulcer can lead to 1.5–2 million new cases of corneal blindness annually according to the World Health Organization (WHO) reports [7]. With this background our aim of this study to identify the etiological agents causing corneal ulcer presenting in our tertiary care hospital.

Materials and Methods:

This is a Prospective study conducted at Narayan medical college and hospital (NMCH) Jamuhar Sasaram, Bihar over a period of 1-year 3 months from January 2019 to March 2020. Before the commencement of the study Institutional Ethics Committee (IEC) clearance was obtained. A total of 61 patients with a clinically diagnosed corneal ulcer were included in this study. All the Patients with corneal ulcers presented to Outpatient department of Ophthalmology at NMCH. Corneal ulcers which were included in this study was defined as a loss of the corneal epithelium with underlying stromal infiltration and suppuration associated with signs of inflammation with or without hypopyon. A detailed clinical history including symptoms, previous treatment with topical corticosteroids, antibiotics, antifungal and predisposing ocular conditions like trauma with vegetative material, occupations, other associated risk factors, demographic profile and verbal consent were obtained.

All the patients were examined under slit lamp bio microscope and characteristics of ulcer details were noted.[Figure 1A] Before taking corneal scrape, use of topical medications specially antimicrobials were withdrawn at least for 12 hours (preferably for 24 hours). [8]

Corneal scraping was performed under topical anesthesia preferably one drop of 0.5% proparacain in the inferior fornix of the affected eye. Topical proparacain had the least amount of bactericidal action compared to other anesthetic agents such as tetracain and xylocain..[9][10] Corneal scraping was done under strict aseptic conditions by an ophthalmologist using a sterile surgical blade number 15 under magnification of a slit lamp bio microscope. After corneal scrapings were done, the scraped material was inoculated directly in the blood agar for bacterial culture, two Sabouraud Dextrose Agar(SDA) media with antibiotics chloramphenicol for fungal culture and smeared onto two slides, one for Gram staining for bacterial evidence, pus cells and the other for 10% Potassium hydroxide (KOH) mount to look for fungal elements

After doing the corneal scraping, the patients were treated with antibiotic eye drops and/or antifungal eye drops depending upon KOH and Gram' staining reports. The patients were admitted or examined daily if feasible with a slit lamp so that their response to treatment can be evaluated and the frequency of antibiotic and antifungal can be titrated. [11] The anti-microbial therapy and follow up visits can be revised based on culture and sensitivity report.

Inoculated blood agar were incubated aerobically at 37 °C which were discarded if no growth was observed after 48 hours of incubation. Whereas SDA for fungal cultures[Figure 1 B] were incubated at 25 °Cat BOD and 37°C incubator and examined daily, and discarded at 3 weeks if no growth was present.

The specific identification of bacterial pathogens was done as per standard protocols. Fungi were identified by their colony characteristics on SDA and cellular morphology with the help lacto phenol cotton blue stain.[Figure 1C]

Result:

Of 61 patients included in study male to female ratio is almost equal (30 males and 31 females) and the age range of 41–60 years were the most commonly affected group[Fig2]. Among total sixty-one patients, forty-seven (77%) were agricultural workers followed by seven homemakers (11%), two professionals (3%), four students (7%) and one others. [Fig3]

The common risk factors in our study were trauma with vegetative materials in 27(44%) patients while working on agricultural field followed by unknown factors in 13 (21%) patients, animal tail injury in 6 (10%) patients, preexisting ocular diseases in 6 (10%) patients (corneal opacity in 3 patients,postkeratoplasty , trichiasis and chronic dacryosistis one in each patient) and inadvertent steroid use in 5 (8%) patients[Tab;1].

Most of the patient 25 (41%) came to eye OPD within one week of corneal ulcer and 20 (33%) patients reported between 2 to 4 weeks of corneal ulcer , and only 16 (26%) patients reported after a month.[Fig;3]

During presentation to eye OPD 15 (25%) patients had documented history of steroid with or without antibiotics, 26 (43%) has history of application of anti-fungal and or antibacterial eye drops without steroid, 6 (10%) patients had used undocumented eye drops and

only 14 (23%) patients presented without prior use of any medication.[Tab;2]

In our study, 29 (48%) patients were found to be culture positive. While the remaining 32 (52%) patients were found to be culture negative. Among the 29 culture positive patients, 23 (79%) patients were positive for fungi, while 6 (21%) patients were positive for bacteria [Table 3]. Among fungal isolates Fusarium spp. found in 11 (48%) patients followed by Aspergillus spp found in 6 (26%) patients, Bipolaris specifera in two patients and one each of Alternaria spp, urvularia spp, Alternaria spp, Scopulariosis brumptii,and Trichothesium.. Common bacterial isolates were Staphylococcus aureus in four subjects followed by Streptococcus and Pseudomonas aeruginosa one in each patient.[Tab;4]

All Staphylococcus aureus were susceptible to vancomycin followed by chloramphenicol, teicoplanin, linezolid and levofloxacin whereas streptococcus spp was sensitive to all antibiotics. And Pseudomonas aeruginosa was sensitive for Levofloxacin, Meropenem, Tobramycin, Gentamicin, Pipramicin, tazobactum and Ofloxacin.

On analyzing the month wise trend of corneal ulcer cases, majority of cases 24(39%) presented to ophthalmology OPD in winter months specially from October to December. [Figure;4]

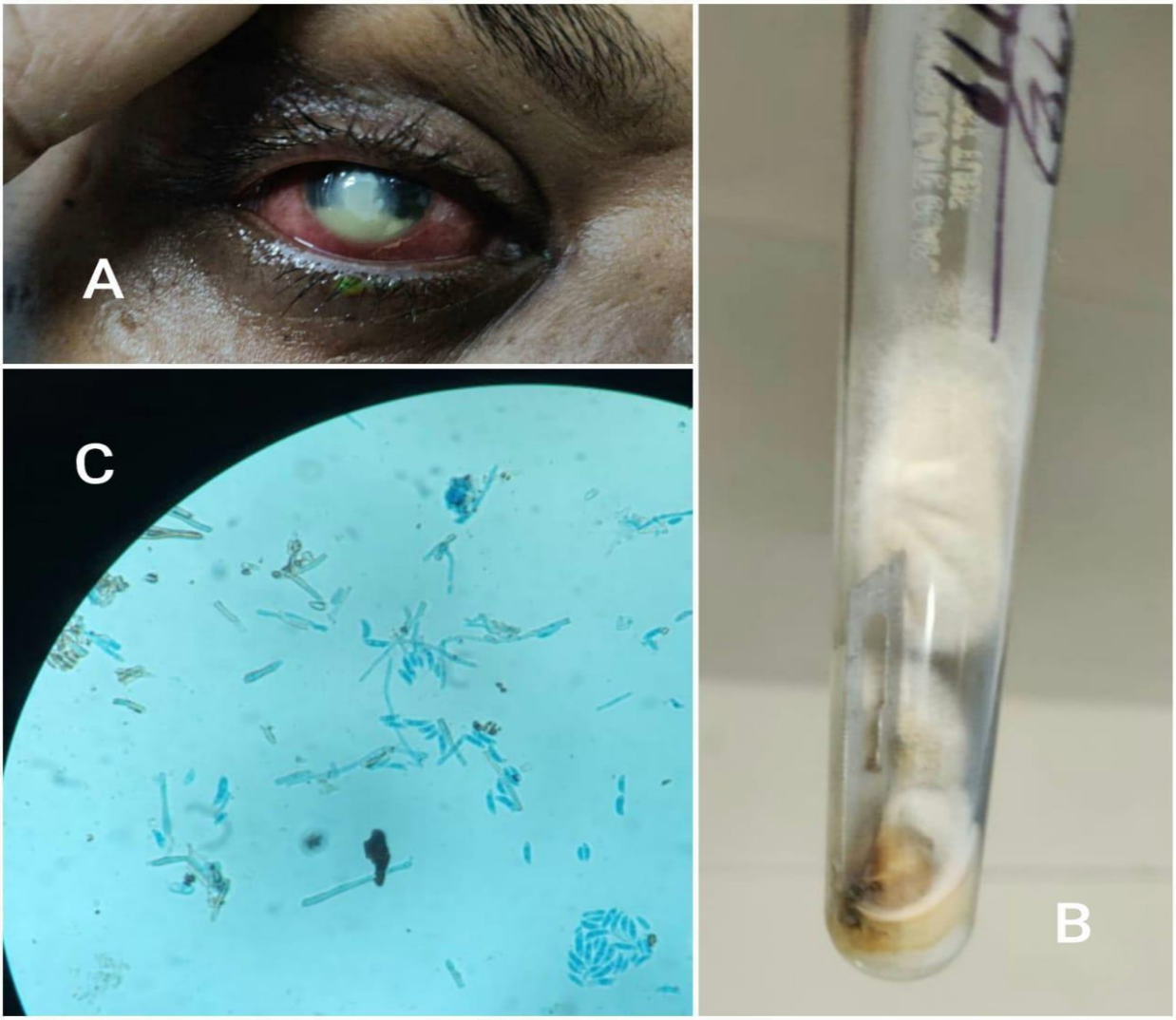

Figure 1: A. Slit lamp picture of Fungal corneal ulcer **B**. Fusarium spp. growth on Sabouraud's Dextrose Agar **C**.Lacto phenol cotton blue wet mount examination of Fusarium spp.

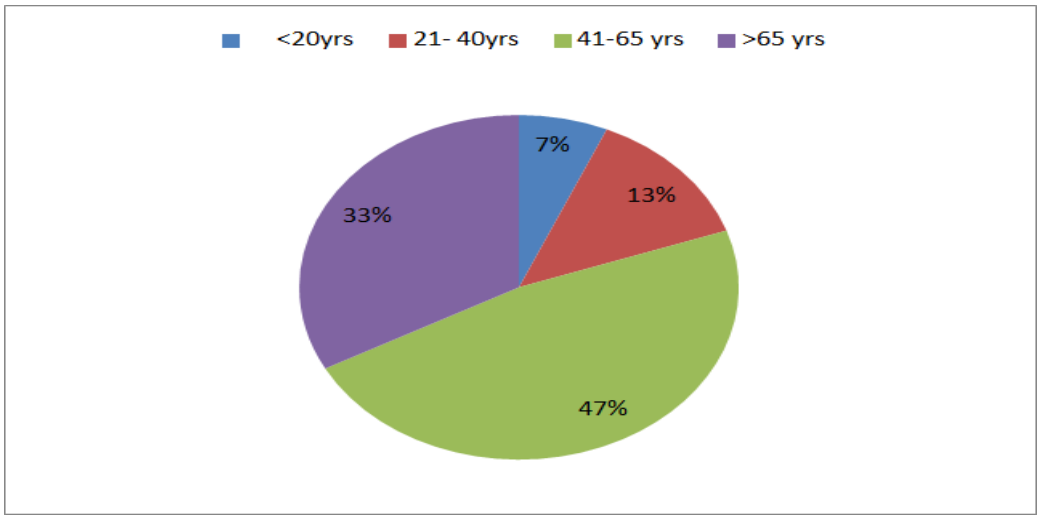

Figure 2: Age groups of patients with microbial keratitis

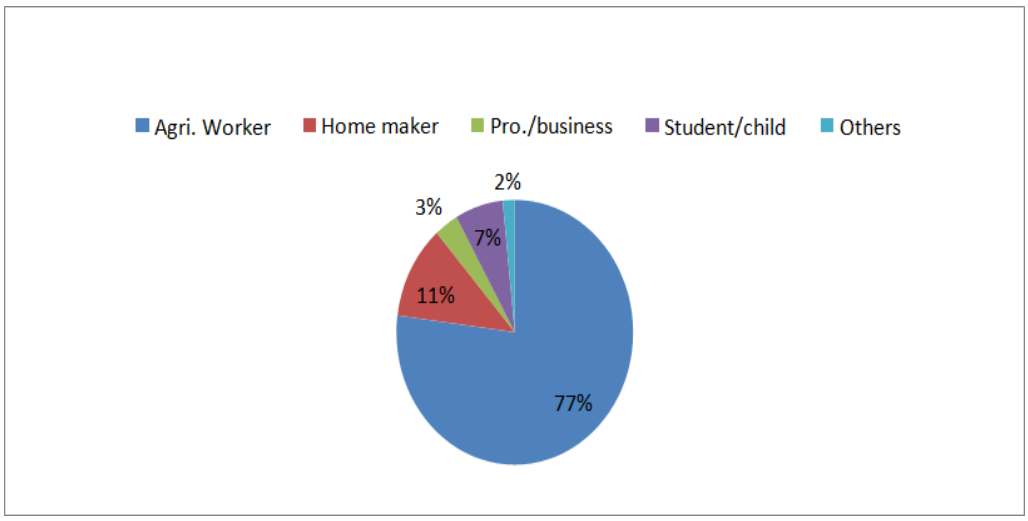

Figure 3: Profession of patients of corneal ulcer.

Table 1: Risk factors associated among the corneal ulcer patients.

Risk factor	**Number (%)**
Vegetative materials	27 (44.3%)
Animal tail	6(9.8%)
Miscellaneous trauma Stone Iron rod Finger nail	 1(1.6%) 1(1.6%) 1(1.6%)
Coexisting ocular diseases Corneal opacity(3), post penetrating keratoplasty(1), trichisis(1),chronic dacrocystitis(1)	6(9.8%)
Coexisting systemic diseases (chemotherapy)	1(1.6%)
Unknown	13(21.3%)
Inadvertent use of steroids	5(8.2%)

Table 2: Treatment History of patients attending outpatient department.

History of prior drugs used			Not used prior drugs
Documented		Un documented	
Steroid with or without antibiotics 15(24.6%)	Non steroidal (antifungal anti bacterial) 26(42.2%)	6(9.8%)	14(22.9%)

Table 3: Growth pattern of microbial agents

NO Growth(32)	Fungal Growth(23)	Bacterial growth(6)
KOH(+):- 10(31.3%) KOH(-):- 22(68.8%)	KOH(+):- 21(91.3%) KOH(-):- 2 (8.7%)	6 Gram's(+):- 4(66.7%) Gram'(-):- 2(33.3%)

Table 4: Microbial agents responsible for corneal ulcer.

	Fungal isolates 23		Bacterial isolates 6	
s.no	Agent	No.(%)	Bacteria	No. (%)
1	Aspergillus spp	6(26.1%)	Staphylococcus aureus	4(66.7%)
2	Fusarium spp	11(47.8%)	Streptococcus spp	1(16.7%)
3	Curvularia lunta	1(4.4%)	Pseudomonas aeruginosa	1(16.7%)
4	Alternaria spp	1(4.4%)		
5	Scopulariops is brumptii	1(4.4%)		
6	Trichothesium	1(4.4%)		
7	Bipolaris specifera	2(8.8%)		

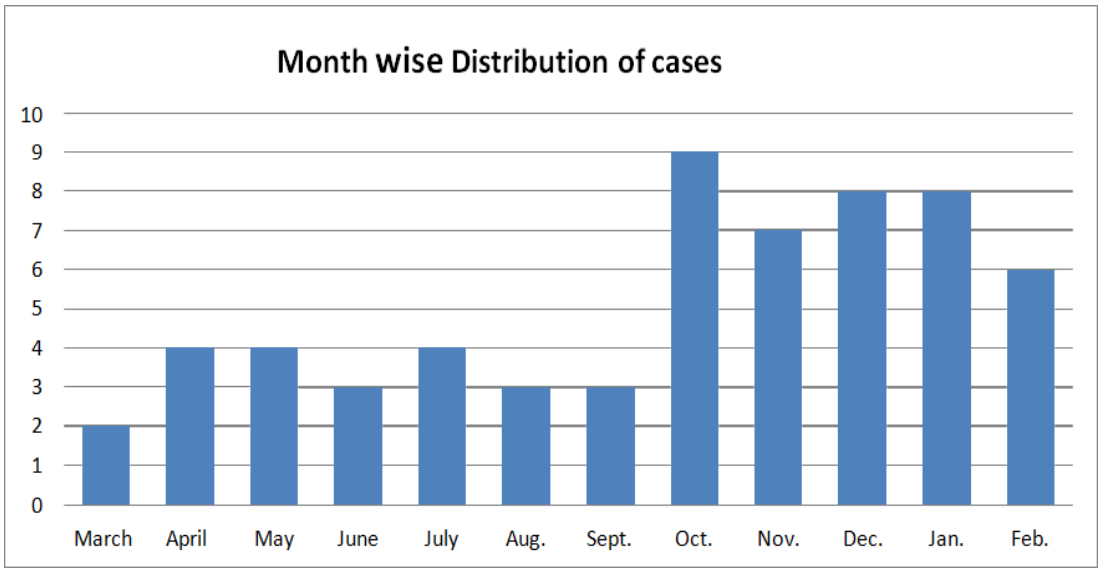

Figure 4: Seasonal variation of corneal ulcer cases.

Discussion:

Corneal ulcer is a major cause of prolonged ocular morbidity and loss of vision. Proper management and treatment of corneal ulcers depends upon identification of the etiological agents, so that an appropriate antimicrobial agent can be administered on time to prevent blindness. [5] In our study we have found almost equal distribution among both the sexes which is similar to a study conducted by Upadhyay *et al.* in Nepal [12]. This could be because of agricultural land and low socioeconomic state where both male and female go to field for work as daily wages laborers. [9] According to study conducted by Titiyal *et al.*[13] and Shubhra Mehta [14] where they have found male were more affected compare to female.

In our study, 29 (48 %) out of 61 patients belonged to age group 41-65 years. A similar study by Li *et al.* also noted that the age group which is highly presented as corneal ulcer belongs to age group 50-59 years, accounting for 83.21%. [15] and Sharmila Suwalet.al. also reported 40% of their corneal ulcer belongs to age group of 51 to 60 years of age.[16] in contrast to Rumpa Sahafrom new Delhi Showed most affected age group was 31-40 yr. [17] Among 61 patient in our study 47 (77%) belongs to agricultural workers which accounts more prevalence, this could be because of our tertiary care center serves more of rural population and belonging to lower socioeconomic status they are mostly engaged in outdoor field work which is similar to study conducted by Mudhol R et al [18] in contrast to Hagan Met.al. who reported 16% of agricultural population reported with corneal ulcer. [19] Ocular injury in 36 (59%) cases was the most common predisposing factor of which 27 (44%) cases of trauma was from vegetative matter while working on field followed by 6 (10%) cases of trauma by animal tail and 3 (5%) cases of other form of trauma whereas 13 (21%) patients presented with unknown risk factors for corneal ulcer which is in concordance with study of Laltanpuia Chhangteet.al**.** conducted at Haldwani also reported ocular injury in 72.4% of cases was the most common predisposing factor [20] Other studies by Assudani *et al.* also Trauma was found to be the major predisposing cause of corneal Ulcer.[21]

In our study we have noticed that 47 (77%) patients already had used some form of topical medications before coming to Eye OPD. Out of that 26 (42.62%) had used nonsteroidal eye drops (antifungal and or antibacterial eye drop) and 15 (24.59%) had used steroidal eye drop and 6 (10%) had used undocumented eye drop whereas 14(23%) patients reported to eye department without use of any medication. This high use of topical medicine before coming to hospital may be due to easy availability of medicine from chemist or some quack suggestion. Rekha mudhol et.al. were also reported 62% of patient had used topical application of medicine before visiting to hospital [4]. A study by Mc Donnel et al. also found that 50% of patients with corneal ulcer got treated with antibiotics before culture report. [22] The lower value of culture positive scraping (47.5%) in our case may be due to high uses of antimicrobial agents.

In this study, 47.5% of the corneal scrapings were culture positive whereas Gonzales et.al.[23] and Laltanpuia et al.[20} showed 57.3% and 68.4% respectively. Out of 47.5% culture positive 37.7 % were fungal isolates and 9.8 % were bacterial isolates whereas a similar study conducted by Bharathi *et al.*[24] who showed 44.1% corneal scraping were positive for fungal culture. In the present study among 37.7% of fungal growth 91.3% were correlating with 10% KOH report which is in accordance with study of Vajpayee RB et al who had reported KOH sensitivity of 94% [25] and we also report 66.6% of bacterial growth were correlating with grams' stain findings in similar to Williams G et al [26] who reported gram stain sensitivity of 63%.

Among the fungal isolates in our study Fusarium spp 11(47.8%) was the most isolated species followed by Aspergillus spp. 6(26%), Bipolaris specifera 2(8.6%) and one species each of Curvularia lunta, Alternaria spp, Scopulariopsis brumptii and Trichothesium. In contrast to Alkatan et al. [27] study who had reported aspergillus spp 27.6% as higher incidence than Fusarium spp. 17.2%. Comparable results were obtained in studies done by Idiculla et al revealing *Fusarium spp* (50%) and *Aspergillus spp* (34.4%) predominate fungal isolates. [28]

The difference in the isolation rates of these fungal pathogens can be explained by the difference in the climate and the natural environment of individual regions. Studies in the South Indian region have shown a higher incidence of *Fusarium* as compared to studies in the northern or western India. [29]

In our study Bacterial corneal ulcer were predominant by Staphylococcus aureus accounting 4(66.6%) subjects followed by one each of Streptococcus spp and Pseudomonas aeruginosa which is comparable with study of Das et al. [30] who also found the most common bacterial isolate was S.aureus followed by Pseudomonas.

After doing the corneal scraping, the patients were treated with antibiotic eye drops and/or antifungal eye drops depending upon KOH and Gram' staining reports. The patients were admitted or examined daily if feasible with a slit lamp so that their response to treatment can be evaluated and the frequency of

antibiotic and antifungal can be titrated. [11] The anti-microbial therapy and follow up visits can be revised based on culture and sensitivity report. . All the patients were responded well during the treatment and no complications were noted

The summer month have higher frequency of infectious keratitis and P.aeruginosa positivity in a study by Mathew Gorski et al.[31].A significant seasonal variation in the frequency of one organism,Pseudomonas aeruginosa was identified(p= <0.0001); upto 47.6% of culture positive ulcer in the summer were P.aeruginosa positive, whereas culture in the remaining seasons were 0%, 9.1% and 12.5% positive . 44.5% ulcers presented in the summer,12.3% in the fall,21.9% in the winter and 21.3% in the spring (p<0.0001%).Possible factors leading to this increased summer presentation include warmer temperature, higher humidity and greater ocular exposure to water. Retrospective analysis by Charles C.Lin et al [32] revealed an uneven distribution of fungal keratitis throughout the year (p<0.001) with peaks in July and January. A higher incidence of fungal keratitis occurred during the months (July and January) corresponding to the windy and harvest season, during which time infection from vegetative corneal injury may be more likely.

No significant seasonal trend was observed for the combined bacterial keratitis group. The predominant fungal organism was Fusarium spp (42.3%) and the predominant bacterial organisms were Streptococcus pneumoniae (35.1%), Pseudomonas aeruginosa (24.3%), and Nocardia spp (8.1%).[32].

In our study the peak (39.34%) in the incidence of corneal ulcer cases occurred in winter (October –December) that correspond to harvesting season of Kharif crops that is mainly rice(paddy) in the region of south west Bihar[Figure-4].

Clinician should increase their vigilance and education to high risk patients during these periods and potentially modify empirical treatment requirement.Protective devices like goggles with side cover(Chatterjee et al) [33] and prompt screening of any ocular injury during harvesting season may prevent from corneal ulcer and its sequel like corneal blindness.

Conclusion:

In our study we found that in the region of South West Bihar, fungal corneal ulcer specially Fusarium spp were the most common organism responsible followed by bacterial ulcers due to S. aureus and the most common season was during harvesting of paddy crop in winter. Protective devices like goggles with side cover during harvesting of crops with timely and appropriate antimicrobial therapy will help to prevent the cases of corneal ulcer and its sequel.

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Conflict of Interest: Nil
Source of support:Nil