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Original Research Article.

Evaluation of Microbiological Profile of Corneal Ulcer Cases Diagnosed at a Tertiary Care Hospital

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ABSTRACT

Background: Corneal infection is a leading cause of ocular morbidity and blindness worldwide.[1] Trachoma alone affects more than 500 million people, of which six to nine million are blind. Herpes Simplex Virus type 1 (HSV-1) is the most common cause of corneal ulcer but other etiological agents frequently associated with corneal ulcer include bacteria (Staphylococcus aureus, Staphylococcus epidermidis, Streptococcus pneumoniae, Streptococcus pyogenes, Moraxella species, Pseudomonas aeruginosa, Proteus species, Klebsiella pneumoniae, Yersinia species and Escherichia coli), fungus (Candida albicans, Aspergillus flavus, Fusarium solani, Penicilium species and Aspergillus fumigates) and parasites (Acanthamoeba). Hence; the present study was undertaken for assessing microbiological profile of corneal ulcer cases diagnosed in a tertiary care.

Materials and Methods: The present study was conducted in the Department of Microbiology, Chalmeda AnandRao Institute of Medical Sciences, Karimnagar, Telangana, India. For this study, patients with suspected microbial corneal ulcers were included and their socio-demographic data and risk factors were recorded. A corneal ulcer was defined as a corneal infiltrate associated with an overlying epithelial defect. Corneal scrapings were routinely collected from patients with corneal ulceration by an ophthalmologist viewing through a slit lamp. Scrapings were taken from the edge of the ulcer before administration of any antimicrobials using a sterile #15 blade after instillation of topical 4% xylocaine.

Results: S. aureus was most commonly seen in conjunction with other microorganisms in mixed infection. Strep. Pneumoniae is the only seen with Fusarium. Table 2 shows number and percentage of type of isolates. Only bacteria were seen in 17 samples, 18 samples had only fungi, mixed microorganisms are seen in 5 cases.

Conclusion: Routine microbiological examination of patients with corneal ulcer is necessary in order to analyze and compare the changing trends of the etiology and their susceptibility patterns which would be beneficial in applying an appropriate antimicrobial treatment.

Keywords: Corneal Ulcer, Microbiological, Acanthamoeba.

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INTRODUCTION

Corneal infection is a leading cause of ocular morbidity and blindness worldwide.[1] Trachoma alone affects more than 500 million people, of which six to nine million are blind. Herpes Simplex Virus type 1 (HSV-1) is the most common cause of corneal ulcer but other etiological agents frequently associated with corneal ulcer include bacteria (Staphylococcus aureus, Staphylococcus epidermidis, Streptococcus pneumoniae, Streptococcus pyogenes, Moraxella species, Pseudomonas aeruginosa, Proteus species, Klebsiella pneumoniae, Yersinia species and Escherichia coli), fungus (Candida albicans, Aspergillus flavus, Fusarium solani, Penicilium species and Aspergillus fumigates) and parasites (Acanthamoeba).[2,-4]

In addition, Pseudomonas a Gram negative opportunistic bacteria is also commonly associated with keratitis arising from contact lens wear, which ultimately leads to corneal ulcer. These pathogens lead to corneal damage directly or by release of toxins and enzymes or by activating the host immune system.[5] An intact corneal epithelium acts as a barrier for the majority of microorganisms. Microorganisms can penetrate through a breach in the epithelium either due to penetrating or perforating ocular trauma or due to surgery.[6]

Hence, the present study was conducted for evaluation of microbiological profile of corneal ulcer cases diagnosed at a tertiary care hospital.

MATERIALS AND METHODS

The present study was conducted in the Department of Microbiology, Chalmeda AnandRao Institute of Medical Sciences, Karimnagar, Telangana, India. The ethical clearance for the study was approved by the ethical committee of the hospital. For this study, patients with suspected microbial corneal ulcers were included and their socio-demographic data and risk factors were recorded. A corneal ulcer was defined as a corneal infiltrate associated with an overlying epithelial defect. Corneal scrapings were routinely collected from patients with corneal ulceration by an ophthalmologist viewing through a slit lamp. Scrapings were taken from the edge of the ulcer before administration of any antimicrobials using a sterile #15 blade after instillation of topical 4% xylocaine. Direct microscopy was performed by taking the scrapings on two glass slides, one for Grams staining and the other for KOH mount. To prepare the culture, corneal scrapings or buttons were inoculated bed side on blood agar, chocolate agar, MacConkey agar and Sabouraud's dextrose agar (SDA) in multiple C shaped streaks. After overnight incubation, bacterial culture was confirmed by growth on blood agar, chocolate agar and MacConkey agar followed by standard biochemical tests according to the clinical and laboratory standards institute (CLSI) guidelines. The statistical analysis of the data was done using SPSS version 11.0 for windows. Chi-square and Student's t-test were used for checking the significance of the data. A p-value of 0.05 and lesser was defined to be statistical significant.

Table 1: Number of type of isolates

Type of microorganism	Number of samples
Only bacteria	17
Only fungal	18
Mixed	
Both bacteria	4
Bacteria and fungi	1
Total number of isolates	40

Table 2: Pathogens isolated from patients with mixed infections

Fungi	Bacteria
Fusarium	Strep. Pneumoniae
Curvularia	S. aureus
Fusarium	S. aureus
Aspergillus niger	S. aureus
Hyalohypomycetes	S. aureus

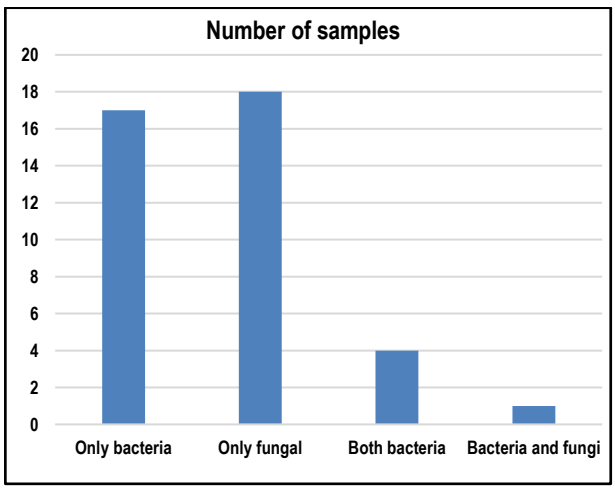

Fig 1: Number of type of isolates

RESULTS

Table 1 shows number and percentage of type of isolates. Only bacteria were seen in 17 samples, 18 samples had only fungi, mixed microorganisms are seen in 5 cases. (Fig 1) Table 2 shows pathogens isolated from patients with mixed infections. S. aureus was most commonly seen in conjunction with other microorganisms in mixed infection. Strep. Pneumoniae is the only seen with Fusarium.

DISCUSSION

In the present study, we observed that microbial etiology was bacterial in 17 and fungal in 18 cases. Five subjects showed mixed growth - both bacterial and fungal growth. Suwal S et al determined the microbiological profile of corneal ulcer cases diagnosed among patients visiting Tilganga Institute of Ophthalmology (TIO), Nepal. A total of 101 corneal scrapping samples were tested for routine culture and antibiotic susceptibility at the pathology department of TIO Nepal from April to October 2014. Microorganisms were identified by using standard microbiological procedures following the manual of American Society for Microbiology (ASM) and their antibiotic susceptibility test, performed by Kirby-Bauer disc diffusion method in conformity with the CLSI guideline. Out of 101 samples analyzed, 44.6% (45/101) showed positive growth with bacterial isolates i.e., 56% (25/45), more prevalent than fungus i.e., 44% (20/45). Among bacteria Streptococcus pneumoniae (31.1%, N = 14) was isolated in highest number whereas Fusarium (13.4%, N = 6) was the most common fungus species. Pseudomonas aeruginosa was the only Gram negative bacteria isolated from corneal ulcer cases. All bacterial isolates were found to be susceptible to the quinolone group of antibiotics (moxifloxacin followed by ofloxacin and ciprofloxacin). These findings showcase the current trend in the microbiological etiology of corneal ulcer in Nepal, which have important public health implications for the treatment as well as prevention of corneal ulceration in the developing world.[6] Feilmeier MR et al reported demographic, microbiological, therapeutic, anatomical, and visual results of corneal ulceration in the elderly patients seen at a tertiary eye care centre in south India. 102 consecutive cases of microbial keratitis in patients 65 years and older were studied. Inclusion criteria were: (i) presence of corneal stromal infiltrate upon slit lamp examination; and (ii) microbiological evaluation of corneal scrapings for suspected microbial keratitis. The principal predisposing factors identified in this study were ocular disease (38.2%), previous ocular surgery in the same eye (29.4%), trauma (17.6%), and severe systemic disease (16.7%). Contact lens wear was associated with only two cases (2.0%). 99 organisms were isolated in cultures of corneal scrapings from 74 (72.5%) of the 102 cases. Staphylococcus epidermidis (31.1%), filamentous fungi (25.7%), and Streptococcus pneumoniae (13.5%) were the most common isolates. 12 eyes (11.8%) required surgery, 15 (14.7%) eventually required evisceration, and nine (9.6%) of the 94 followed patients achieved an unaided vision of 20/60 or better at last follow up. This work represents the largest recent single centre study on (non-viral) microbial keratitis in the elderly, its management, and outcomes of therapy. While the predisposing factors differ from those of general population, the spectrum of microbes responsible for keratitis in the elderly appears to reflect the local microbial flora rather than a predilection for elderly patients. Delay in diagnosis

and systemic conditions associated with advancing age probably contribute to poorer outcome from therapeutic measures.[7]

Ranjini CY et al determined the etiologic diagnosis of infectious corneal ulcers at Tilganga Institute of Ophthalmology, a tertiary teaching hospital in Kathmandu Nepal, from 2006-2009. This study involved a review of all microbiology records at Tilganga Institute of Ophthalmology from August 2006 through July 2009. Microbiologic records from the corneal scrapings of all patients suspected of having infectious corneal ulcers were included. Corneal scrapings were obtained from 468 patients. The average patient age was 52 years, and 55% of the affected cases were males. Microorganisms were grown from 185 of the corneal scrapings (40%). Pure bacterial cultures were obtained from 72 patients (39%), and pure fungal cultures were obtained from 113 patients (61%). Gram stain was 75% sensitive (95% confidence interval, 0.632-0.841) in identifying bacterial infection, whereas KOH prep was 80.5% sensitive (95% confidence interval, 0.718-0.871) in identifying fungal organisms. Of 72 bacterial isolates, 50 isolates (69%) were Streptococcus pneumoniae, the most common organism isolated in this study. Of 113 fungal isolates, 40 of isolates (35%) were identified as Aspergillus sp. They concluded that fungal organisms (61%) are the most common cause of infectious keratitis in this patient population. Of all organisms, S. pneumoniae was the most common organism identified. Smear microscopy is reliable in rapidly determining the etiology of the corneal infection and can be used to help guide initial therapy in this setting.[8]

Kunimoto DY et al and Amrutha KB et al identified the prevalence and microbial profile of infectious keratitis in a tertiary eye care hospital, and to test for the in vitro antimicrobial resistance of the bacterial isolates. A total of 312 patients presenting to a tertiary eye care hospital with infected corneal ulcer were enrolled in this study. Their socio-demographic data and risk factors were recorded. Corneal scrapings collected from the edge of the ulcer were processed for direct gram stain and KOH mount. Culture was recovered on blood agar, chocolate agar, MacConkey agar and Sabouraud's dextrose (SDA) agar in multiple C shaped streaks. After overnight incubation, bacterial culture was followed by standard biochemical tests and antimicrobial sensitivity according to the clinical and laboratory standards institute (CLSI) guidelines. Inoculated SDA was inspected daily for up to 10 days and the growth was identified by its colony morphology, pigment production and lacto-phenol cotton blue mount examination. Of 312 patients, a microbial etiology was established in 117 cases (37.5%). Of these, 72 (61.5%) were male. The age range of 41-60 years was the most affected group. Of 117 positive cases, 52 (44.5%) were bacterial, 58 (49.5%) were fungal and 7 (6%) patients showed mixed bacterial and fungal infection. The most common isolated fungus was Fusarium which was detected in 36 (31%) cases, followed by Aspergillus spp in 13 (11%) subjects. Staphylococcus aureus was the most common isolated bacteria. All Gram positive cocci were susceptible to vancomycin followed by gatifloxacin, whereas all Gram negative bacilli were susceptible to gatifloxacin. It was concluded that routine microbiological examination of patients with corneal ulcer is necessary in order to analyze and compare the changing trends of the etiology and their susceptibility patterns.[9,10]

CONCLUSION

Within the limitations of the present study, it can be concluded that routine microbiological examination of patients with corneal ulcer is necessary in order to analyze and compare the changing trends of the etiology and their susceptibility patterns which would be beneficial in applying an appropriate antimicrobial treatment.

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