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Microbiological profile of Ulcerative Keratitis in a tertiary care hospital

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Abstract:

Introduction: Microbial keratitis is a common potentially sight threatening ocular infection that may be caused by bacteria, fungi, viruses or parasites. The concept of ocular pathogen is outmoded. Given the optimal condition any organism can cause keratitis. Microbiological evaluation is crucial in differentiating infectious from non infectious inflammatory conditions of the cornea. **Material & Methods:** 100 consequent suppurative corneal ulcer cases were studied. Corneal scrapings were collected and subjected for examination by Grams stain, 10% KOH, bacterial and fungal culture. Antibiotic susceptibility of the bacterial pathogens was performed by Kirby Bauer disc diffusion method.

Results: Of the 100 patients studied, 77% were from rural areas and 54% presented with ocular trauma. Direct microscopy was positive in 48% and culture in 67% cases. 67.17% were bacterial, 25.37% were fungal, 7.5% polymicrobial and no *Acanthamoeba* were identified. CONS, *S. aureus*, *Ps. aeruginosa* were the common bacterial isolates and *Fusarium spp* was the predominant fungal isolate. Ciprofloxacin was the most effective antibiotic. **Conclusion:** Suppurative keratitis being a sight threatening disorder, early suspicion, rational use of laboratory diagnostic procedures and appropriate therapy can go a long way towards reducing ocular damage.

Key words: CONS; Fusarium; Keratitis; Pseudomonas; Trauma.

Introduction

The problem of blindness is universal but the magnitude is much more in India, having 1/4th of the world's total blind population. Corneal infections are a leading cause of ocular morbidity and blindness worldwide [1]. It is estimated that 6.8 million people who have vision less than 6/60 in at least one eye is due to corneal disease. In India 15.4% of blindness are attributed to corneal diseases and infections constitute the most predominant cause of corneal blindness, second only to cataract [2]. Corneal

ulcerations affect 113 per 100000 populations in India. Surveys conducted in Africa and Asia showed corneal scarring as a major cause of visual disability. It accounts for 20 – 30 % of all blindness in the developing countries [3].

The occurrence rates of corneal pathogens are largely dictated by local microbial flora. While viral infections are the leading cause in developed nations with *Acanthomeba* in contact lens wearers; bacteria, fungi and *Acanthomeba* are important aetiological agents in developing countries [4].

Untreated infective keratitis may result in corneal perforation, with the potential for development of endophthalmitis and may require evisceration. To devise a comprehensive strategy for diagnosis and treatment of corneal ulcers it is of paramount importance to known the regional etiological agents.

Material and Methods

One hundred consequent patients presenting with suppurative corneal ulcers, attending the ophthalmology department were included in the study. All cases of typical viral and non infective ulcers were excluded. After an informed consent a proforma was filled out for each patient documenting age, sex, occupation, residence, predisposing factors and treatment history.

After application of 0.5% proparcaine hydrochloride topical anaesthetic, corneal scrapings were collected by an ophthalmologist under direct visualisation through slit lamp biomicroscope. Purulent material over the ulcer was removed using sterile cotton swab and discarded. Using bent tip of 21 gauge disposable needle multiple corneal scrapings were obtained from the leading edge and base of the ulcer.

Scrapings were smeared onto glass slide, fixed with 95% methanol and stained with Gram stain. From the scrapings 10% KOH mount was done and examined for fungal elements and *Acanthamoeba* cysts. Scrapings were inoculated directly onto blood agar, chocolate agar, MacConkey agar and 2 sets of Sabouraud dextrose agar with chloramphenicol. All bacterial cultures were incubated at 37^0C, with chocolate agar in CO_2 jar, examined at 24 hrs, 48 hrs and 72 hrs. Fungal cultures were incubated at room temperature and 37^oC and incubated up to 6 weeks. Isolates were identified using standard protocol [5].

Cultures were considered significant when [6]:

i) Growth of same organism on 2 or more media

ii) Semi confluent growth on one media consistent with smear findings.

Antimicrobial susceptibility of bacterial isolates to antibiotics used in our setup was done by Kirby-Bauer disc diffusion method against penicillin, tetracycline, chloramphenicol, gentamicin, norfloxacin, ciprofloxacin and cefazolin. Susceptibility testing was not done for *Str. Pneumoniae* and penicillin was not tested for Gram negative organisms.

Results

Among 100 corneal ulcer patients, 61% were males and 39% were females. 77 patients were from rural areas and 23 from urban areas. 34 patients were agricultural workers. Predisposing factors were present in 70 cases; trauma 54(77.14%), local corticosteroids 4(5.71%), chronic dacrocystitis 3(4.29%), and blepharitis, endophalmitis, surgery of same eye and diabetes mellitus in 2(2.86%) each entropion in 1 (1.43%). 69% ulcers were located centrally and 46% had associated hypopyon.

Of the 100 ulcers studied, 48% showed presence of either bacterial or fungal elements on direct microscopy. 67% ulcers were culture positive and in 2 cases *Pneumococci* were seen on Gram stain but did not grow on culture.

Table 1: Microbial flora of corneal ulcer

Growth pattern	**No of patients**	**Percentage (%)**
Single bacterial isolate	45	67.17
Single fungal isolate	17	25.37
Mixed growth	5	7.46
Acanthamoeba	0	00.00
Total	**67**	**100**

Table 2: Bacterial isolates

Bacteria	No. of isolates	Percentage (%)
CONS	15	29.41
S. aureus	11	21.57
Ps. aeruginosa	10	19.61
Str. pneumoniae	4	7.85
E. faecalis	3	5.88
Corynebacterium spp.	3	5.88
Acinetobacter spp	2	3.92
B. cereus	1	1.96
Esch. coli	1	1.96
Alkaligenes faecalis	1	1.96
Total	**51**	**100**

Table 3: Fungal isolates

Fungi isolated	No. of isolates	Percentage (%)
Fusarium spp	13	61.91
Aspergillus spp	4	19.05
i) Aspergillus fumigates	2	9.52
ii) Aspergillus flavus	1	4.76
iii) Aspergillus niger	1	4.76
Acremonium spp	2	9.52
Scopulariopsis spp	1	4.76
Curvularia spp	1	4.76
Total	**21**	**100**

Table 2: Bacterial isolates

51 bacterial isolates were obtained from 50 ulcers. One patient had mixed infection of CONS and *Ps.aeruginosa*.

The most effective antibiotic was ciprofloxacin with sensitivity of 91.5% followed by gentamicin 63.8% and cefazolin 61.7%.

Discussion

In the present study an attempt was made to know the causative agents of ulcerative keratitis prevalent in our area with antimicrobial susceptibility of bacterial isolates.

As there is no definite pathogonomic clinical feature, it is difficult to establish the aetiology of corneal ulcer merely on the basis of clinical features. Hence microbiological evaluation is a must in order to attain a definitive diagnosis and ensure specific therapy [7].

The avascular cornea is particularly susceptible to bacterial infections and may perforate in less than 24 hrs if not treated promptly. With an advanced infection or a severe host response, devastating complications like corneal thinning, perforation, scleral extension may occur, that require surgical intervention [8]. Aggressive initial treatment can minimise corneal scars and complications. Corneal ulceration has been recognised as a silent epidemic in developing countries, especially the South-east Asian region [2]. It is predicted that the number of corneal blind people in India will increase to 10.6 billion by 2020 [3].

In the present study 61% were males and 39% were females. 77% patients were from rural areas and 34% patients were agricultural workers. In our country, majority of population is involved in agricultural work and various types of manual labour. Thus they become more prone to injuries, exposure to dust & wind along with poor ocular hygiene, promote development of microbial keratitis. As reported by Gopinathan et al, patients with agriculture-based activities were at 1.33-times greater risk of developing microbial keratitis [9].

Predisposing factors were present in 70 cases with trauma 54(77.14%) being the commonest predisposing factor and local corticosteroid use was seen in 4(5.71%) patients. 46% had associated hypopyon. Trauma is a major risk factor for corneal infection in developing countries. In Paraguay, the percentage of cases with preceding trauma was 48%;

in Madurai, South India 65% and in Eastern India 83% [7].

Of the 100 ulcers studied, 48% showed presence of either bacterial of fungal elements on direct microscopy. 67% ulcers were culture positive and in 2 cases *Pneumococci* were seen on Gram stain but culture negative. Bacteria were isolated from 45 (67.17%) ulcers and fungi in 17(25.37%), 5 (7.46%) patients had polymicrobial infection, of which 4 had bacterial and fungal co-infection. Similar pattern of microbial keratitis was reported by Gopinathan et al; 51.9% bacterial and 38.2% fungal, 2.4% *Acanthamoeba* and 7.5% polymicrobial etiology [9]. *Acanthamoeba* were not detected in our study. However reports from north Karnataka have reported 41.0% fungal and 32.5% bacterial etiology [2].

The commonest bacterial isolates were CONS 29.41%, followed by *S. aureus* 21.57% and *Ps.aeruginosa*19.61%. *Fusarium spp* 61.91% was the predominant fungal isolate. Many studies have reported the common bacterial causes of microbial keratitis as *Ps. aeruginosa*, *S. aureus*, *S. epidermidis*. Gopinathan et al have reported *S. epidermidis* as the predominant bacterial pathogen [9]. Bacterial keratitis caused by *Ps.aeruginosa* is more fulminant and associated with a worse visual prognosis than that caused by most other common bacterial pathogens. *Pseudomonas*-related keratitis can be associated with permanent central corneal scarring and/or irregular astigmatism and visual loss despite optimal management [10]. The most effective antibiotic in our study was ciprofloxacin with sensitivity of 91.5% followed by gentamicin 63.8% and cefazolin 61.7%. Jeng, commented on the emerging resistance of bacterial infections to fluoroquinolones. In addition to changes in resistance patterns, studies have also demonstrated changing patterns of causative organisms over time in a given geographical location [7]. Fungal keratitis account for approximately 28% of ulcerative keratitis, ranging from 6% to 53%, and fungal keratitis is about 50% in culture proven cases. Upto 20% of fungal keratitis are complicated by bacterial coinfection. In our study there were 4 cases of bacterial and fungal coinfection. Filamentous fungi are frequent causes of corneal ulcers. Filamentous agents vary depending on the geography, and climate of each region. *Aspergillus spp* was the most common isolate of fungal keratitis reported in India, South Iran, Nepal and Bangladesh. *Fusarium spp* was found to be the most common cause of fungal keratitis in South India, but the prevalence in other parts of the world is limited [7,2].

Fusarium keratitis has a more aggressive course and is less responsive to treatment than *Aspergillus*. The dematiaecae are of low virulence and produce protracted infections. Rapid diagnosis and proper treatment of fungal keratitis are important as many people require several months of treatment and debridement of the ulcer before instillation of local antifungal [11].

Conclusion

Delicate balance exists between cornea and surrounding environment that helps cornea maintain its integrity in-spite of continuous exposure to pathogens. Corneal ulceration may result when the balance is disrupted and defence mechanisms compromised. Early detection and appropriate treatment are important to minimize permanent visual loss. Patients with risk factors predisposing to microbial keratitis should be educated about their relative risk, be acquainted with signs and symptoms of infection, and advised to consult an ophthalmologist promptly if they experience such warning signs or symptoms.

Monitoring patterns of corneal pathogens, their antibiotic susceptibility can form basis for accurate management and empirical therapy.

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