Screening of Bacterial and Fungal Pathogens in Kerato-Conjunctivitis

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Abstract- *Bacterial conjunctivitis occurs in persons of all races, although differences in frequencies may be reflected by geographical variations of pathogen prevalence. The study was therefore taken up to detect the prevalence of bacterial and fungal pathogens causing occular infections and to study their antibiotic resistant profiles.A total of 44 kerato-conjunctivitis samples were collected, out of which, 31/44 (73%) were fungal isolates. The prevalence of fungal isolates was as follows- 7 Aspergillus fumigatus (22.5%), 3 Aspergillus flavus (9.67%), 4 Aspergillus nidulans (12.9%), 7 Aspergillus niger (22.5%), 10 Fusarium sps. (32.25%).A total of 39/44 (86.36%) bacterial isolates were obtained in this study. The prevalence of bacterial isolates was as follows- 18 Staphylococcus aureus (46.15%), 10 Staphylococcus epidermidis (25.64%), 1 Escherichia coli (2.56%), 5 Pseudomonas aeruginosa (12.82%), 4 Klebsiella pneumonia (10.26%),1 Nocardia sps. (2.56%).*

Keywords- Eye infection, Kerato-conjunctivitis, Bacteria, Fungi, Antibiogram.

I. INTRODUCTION

Infection of the eye leads to conjunctivitis, keratitis, endopthalmitis and other infections which are responsible for increased incidence of morbidity and blindness worldwide.Suppurative keratitis can cause corneal opacity and perforation, which leads to severe visual loss and is the second most common cause for blindness in developing countries. The etiological cause for suppurative keratitis may vary at different geographical locations. Different types of bacteria and fungi that are the important etiological agents affecting cornea orbit and other ocular structures. Fungal infection is a life threatening condition which needs early diagnosis and treatment to save the patient'seye [1].

The present study was taken up to screen for the presence of bacterial and fungal pathogens in kerato-conjunctivitis and to determine the antibiotic sensitivity pattern of the bacterial isolates.

II. MATERIALS AND METHODS

Collection of sample

The kerto-conjunctival samples were collected from different age group of 12-80 years. The samples were collected by rolling a sterile cotton swab over the lower conjunctival fornix. The swab was gently rolled over the entire surface of the kerato-conjunctiva. Care should be taken in order to avoid touching the eye lashes, lid margins (or) fingers with the swab.

Processing of the samples:

Kerato-conjunctival swabs were collected aseptically from the base and edge of the eye from each patient.The swab was inoculated onto blood agar, MacConkey agar and nutrient agar, mannitol salt agar and Sabouraud's dextrose agar and was incubated at 37^{o}C for 24 hrs for bacterial culture and at room temperature for fungal isolates.Next day the colonies were picked up and preliminary identification was done and the bacterial isolates were identified based on standard protocols.LPCB was done for the identification of fungal isolates after 48 to 72 hrs.

III. ANTIBIOTIC SENSITIVITYTEST

Antibiotic sensitivity test of the bacterial isolates was determined by the Kirby-Bauer disc diffusion method[2]. The following were the antibiotics used for the study-amikacin (AK 30), nalidixic acid (NA 30), erythromycin (E 15), vancomycin (VA 30), tetracycline (TE 30), cefoxitin (CX 30), rifampicin (RIF 5) ciprofloxacin (Cip 5), ceftazidime (CAZ 30), cefotaxime (CTX 30), cepifime (Cpm 30), and cefoperazone (CPZ 75).

IV. RESULT

A total of 44 kerato-conjunctivitis samples were collected, out of which, 31/44 (73%) fungal isolates were isolated. The prevalence of fungal isolates was as follows- 7

Aspergillus fumigatus (22.5%), 3 *Aspergillus flavus* (9.67%), 4 *Aspergillus nidulans* (12.9%), 7 *Aspergillus niger* (22.5%), 10 *Fusarium sps.* (32.25%).(Table 1)

Table 1: Percentage of fungal isolates from Kerato-Conjunctivitis

S.NO	FUNGAL ISOLATES	NUMBER	PERCENTAGE (%)
1	*Fusarium sps.*	10	32.25%
2	*Aspergillus fumigates*	7	22.50%
3	*Aspergillus niger*	7	22.50%
4	*Aspergillus nidulans*	4	12.9%
5	*Aspergillus flavus*	3	9.67%

A total of 39/44 (86.36%) bacterial isolates were obtained in the study. The prevalence of bacterial isolates was as follows- 18 *Staphylococcus aureus* (46.15%), 10 *Staphylococcus epidermidis* (25.64%), 1 *Escherichia coli* (2.63%), 5 *Pseudomonas aeruginosa* (12.82%), 4 *Klebsiella pneumoniae*(10.26%),1 Nocardiasps. (2.56%). (Figure 2 & Table 2)

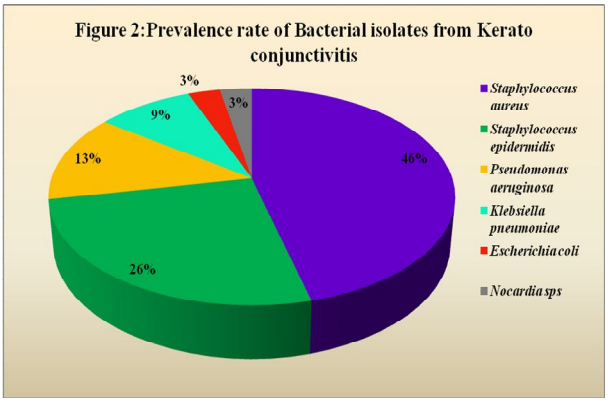

Table 2:Percentage of bacterial isolates from kerato-conjunctivitis

S.NO	BACTERIAL ISOLATES	NUMBER (N)	PERCENTAGE (%)
1	*Staphylococcus aureus*	18	46.15%
2	*Staphylococcus epidermidis*	10	25.64%
3	*Pseudomonas aeruginosa*	5	12.82%
4	*Klebsiella pneumoniae*	4	10.26%
5	*Escherichia coli*	1	2.56%
6	*Nocardia sps.*	1	2.56%

V. ANTIBIOTIC SENSITIVITY TESTING

Staphylococcus aureus isolates were found to be highly sensitive to amikacin and rifampicin(100%) followed by cefoxitin and tetracycline (89%), nalidixic acid (84%) and erythromycin (28%). They were found to be highly resistantto nalidixic acid, tetracycline and cefoxitin (11%). They showed intermediate resistance towards erythromycin (72%) and nalidixic acid (5%).

Staphylococcus epidermidis isolates were found to be highly sensitive to nalidixic (80%) followed by amikacin(70%), cefoxitin and rifampicin (50%) and tetracycline (30%). They were found to be highly resistant to erythromycin followed by cefoxitin and rifampicin tetracycline (20%). They showed intermediate resistance towards erythromycin, tetracycline (30%) and nalidixic acid (10%). Staphylococcal isolates showed 100% sensitivity towards vancomycin.

Pseudomonas aeruginosa isolates were found to be highly sensitive to amikacin, cefipime, ciprofloxacin and ceftazidime (100%) followed by cefoparazone (80%). They were found to be highly resistant to cefotaxime (100%). They showed intermediate resistance towards cefoparazone (20%).

Klebsiella pneumoniae isolates were found to be highly sensitive to amikacin, cefipime, ceftazidime, cefotaxime and cefoperazone (100%) followed by ciprofloxacin (50%).*Escherichia coli* isolates were found to be highly sensitive to amikacin, cefoperazone and cefipime (100%). They were found to be highly resistant to ciprofloxacin, cefotaxime (100%). They showed intermediate resistance towards ceftazidime.

VI. DISCUSSION

Infective keratitis may be caused by bacteria, fungi, protozoa and virus, and the spectrum of microbial pathogens causing keratitis varies according to the geographical locations and climate[3],[4]. Infective keratitis rarely occurs in normal eyes without any predisposing factors. The ocular surface is normally protected from microbial invasion through an intricate anatomic relationship between the cornea, conjunctiva, lacrimal secretory apparatus and precorneal tear film and the eye lids [5]. Any disruption of the same may results in less effective defence against infection and such risk factors may vary with occupation. An association has been shown between the type of risk factor and the microbial aetiology for infection[6]. Corneal injury due to vegetative matter predispose mainly to fungal keratitis, while use of

contact lenses and other non-traumatic risk factors to bacterial keratitis[7]. Thus, the analysis of such associated risk factors and the isolated microbial aetiology will help in understanding the relationship between risk factors and microbial keratitis, and is essential for initiation of empirical antimicrobial therapy with subjective interpretation of presenting clinical features for practicing ophthalmologist.

The aetiology and epidemiology of corneal ulcers vary with the patient population, geographic location and climate, and tends to vary somewhat over time [8],[9]. In the present study, a total of 44 kerato-conjunctivitis samples were collected, out of which, 31/44 (73%) fungal isolates were isolated. The prevalence of fungal isolates was as follows- 7 *Aspergillus fumigatus* (22.5%), 3 *Aspergillus flavus* (9.67%), 4 *Aspergillus nidulans* (12.9%), 7 *Aspergillus niger* (22.5%), 10 *Fusarium sps.* (32.25%). Among the fungal isolates, *Fusarium* (32%) was found to be predominant.

In the present study, a total of 39/44 (86.36%) bacterial isolates were. The prevalence of bacterial isolates was as follows- 18 *Staphylococcus aureus* (46.15%), 10 *Staphylococcus epidermidis* (25.64%), 1 *Escherichia coli* (2.63%), 5 *Pseudomonas aeruginosa* (12.82%), 4 *Klebsiella pneumonia* (10.26%), Nocardia sps. (2.56%).This was in agreement with the studies done in South India [10], [11], Taiwan[12], Thailand[13] and Ghana[14], where *S.aureus*, *Pseudomonas* and *Fusarium* species are the most common causes of bacterial and fungal keratitis respectively. *Pseudomonas aeruginosa* are also more likely to be isolated from contact lens-related keratitis in areas with higher maximum and minimum temperatures [15].

The relative prevalence of mycotic keratitis in Eastern India is lower than southern, western and north-eastern India but higher than Northern India, however, *Aspergillus* and *Fusarium* are the predominant genera associated with fungal keratitis across India. The response to medical treatment is poor in patients with late presentation[16].

Since 1991, Fluoroquinolones (Ciprofloxacin, Ofloxacin, and Levofloxacin) have been available for the treatment of bacterial kerato- conjunctivitis. These are bactericidal and have a broad spectrum of activity against both gram negative and gram positive bacteria. They are most effective against *Staphylococcus aureus and Pseudomonas aeruginosa* [17]. In our study, antibiotic sensitivity test was done for all the bacterial isolates. In which, gram positive isolates were more sensitive to amikacin, rifampicin and vancomycin. The gram negative isolates were more sensitive to amikacin, cefipime and cefoperazone.

Staphylococcus aureus isolates were found to be highly sensitive to amikacin and rifampicin (100%) followed by cefoxitin and tetracycline (89%), nalidixic acid (84%) and erythromycin (28%). They were found to be highly resistant nalidixic acid, tetracycline and cefoxitin (11%). They showed intermediate resistance towards erythromycin (72%) and nalidixic acid (5%).

Staphylococcus epidermidis isolates were found to be highly sensitive to nalidixic (80%) followed by amikacin (70%), cefoxitin and rifampicin (50%) and tetracycline (30%). They were found to be highly resistant to erythromycin followed by cefoxitin and rifampicin tetracycline (20%). They showed intermediate resistance towards erythromycin, tetracycline (30%) and nalidixic acid (10%). Staphylococcal isolates showed 100% sensitivity towards vancomycin.

P.aeruginosa were found to be highly sensitive to amikacin, cefipime, ciprofloxacin and ceftazidime (100%) followed by cefoparazone (80%). They were found to be highly resistant to cefotaxime (100%). They showed intermediate resistance towards cefoparazone (20%).*K. pneumoniae* were found to be highly sensitive to amikacin, cefipime, ceftazidime, cefotaxime and cefoperazone (100%) followed by ciprofloxacin (50%). They showed intermediate resistance towards cefoperazone (50%).*E.coli* were found to be highly sensitive to amikacin, cefoperazone and cefipime (100%). They were found to be highly resistant to ciprofloxacin, cefotaxime (100%). They showed intermediate resistance towards ceftazidime (100%).

VII. CONCLUSION

In our study both the bacterial and fungal pathogens were isolated and most of them exhibited drug resistance. This could lead to prolonged treatment. Microbiology, culture and sensitivity testing remains the gold standard for the identification of pathogens causing microbial keratitis.Suppurative keratitis continues to be a cause of concern and is a major cause of treatable blindness. The microorganisms play a pivotal role as etiological agent thus helping us to have a degree of clinical suspicion for the bacterial and fungal organisms in starting the appropriate initial treatment in keratitis.

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