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Original Research Article

A Study on the Microbiological Profile of External Ocular Infection in a Tertiary Care Center

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Abstract:

Objectives: The present study was to evaluate the microbiological profile and antibiotic sensitivity pattern in patients with external ocular infection.

Methods: After detailed ocular examinations, using standard techniques, specimens for culture and smear were obtained by scraping and swabbing the eyelid margin using sterile blade (# 15) on Bard-Parker handle and sterile broth-moistened cotton swabs in case of blepharitis [8,9]. Similarly, specimens were also obtained from scraping the corneal ulcers. Conjunctival cultures were obtained by wiping a broth-moistened swab across the lower conjunctival cul-de-sac in conjunctivitis cases. In corneal scraping the specimen was cultured on Blood agar in the form of 'c' shape streak. Direct microscopic examinations such as 10% Potassium Hydroxide (KOH) wet mounting, Gram-stain, Kinyoun's acid-fast stain were also done. For fungus identification Slide culture method and LPCB staining were used. The antibiotic susceptibility testing was done by the Kirby Bauer disc diffusion method, as per the CLSI guidelines, 2011.

Results: 100 patients of external ocular infections were enrolled. Among them, 59% were males and 41% were females. Most of the patients (45%) were in age group of >60 years. 16% patients were in age group of <15 years. total conjunctival infection was seen in 44(86.27%) patients. Among them, 37(72.55%) cases were conjunctivitis, 6(11.76%) cases were blepharitis and 2(3.92% cases were dacryocystitis. Out of 51 culture cases, keratitis was seen in 7(13.73%) patients. Gram positive cocci was found in 36 patients. And gram-negative bacilli were found in 15 patients on culture. Out of 51 culture, gram positive cocci were 20(39.22%) coagulase negative Staphylococci (CONS) followed by 12(23.53%) Staphylococcus aureus. Gram negative bacilli were (15.69%) Pseudomonas aeruginosa followed by 3(5.88%) Acinetobacter and 2(3.92%) Klebsiella. Total number of organisms isolated from external ocular infections was 65, out of which bacterial isolate were 51(78.46%) and fungal isolates were 14(21.54%). Gram positive isolates were susceptible to Vancomycin 100%, Teicoplanin 100%, Linezolid 100%, Clindamycin 86.11%, and Ciprofloxacin 83.33%. Gram negative organisms were mostly sensitive to Amikacin 100%, Imipenam 100%, Meropenam 100% and ciprofloxacin 93.33%, ofloxacin 93.33%, gentamicin 93.33%.

Conclusions: External ocular infection is predominantly more in old age male population. Conjunctivitis is more common external ocular infection. Most common gram-positive cocci isolates are the Coagulase negative Staphylococci (CONS) and Staphylococcus aureus. Pseudomonas aeruginosa is the common gram-negative bacilli isolates. Gram positive isolates are more susceptible to vancomycin, teicoplanin, linezolid, clindamycin and ciprofloxacin. Gram negative organisms are more sensitive to amikacin, imipenam, meropenam, gentamicin and fluoroquinolone (ciprofloxacin and ofloxacin).

Keywords: External Ocular Infection, Microbiological Profile And Antibiotic Sensitivity Pattern.

Introduction

Bacteria are generally associated with many types of ocular infections such as conjunctivitis, keratitis, endophthalmitis, blepharitis, orbital cellulitis and dacryocystitis manifestations [1]. Conjunctivitis, inflammation of the mucosa of conjunctiva, is the most frequent ocular case with noticeable economic and social burdens [2].

External ocular infections are affecting and leading to vision loss globally [3]. According to the World Health Organization (WHO), 285 million people were visually impaired worldwide. Out of those, 39 million people were blinded by the year 2010. The report also disclosed that more than 90% of the world's visually impaired people live in developing countries, and surprisingly 82% of the visual impairment, including blindness, was preventable

[4]. In Africa, it is estimated that approximately 2.2 million people were blinded due to ocular infection [5]. One report (2015) in Sudan showed that bacterial external ocular infections are significantly prevalent among the pediatrics population and cause more than 65% of morbidity in all cases [6]. Most ocular infections in the world have been treated using commonly known antimicrobials. Due to this, microbial resistance to antimicrobial agents has become increasingly prevalent in ocular infections including systemic infections on a global basis [7, 8]. When an ocular infection does occur, prompt and effective treatment is necessary to prevent damage from both the infection and the immune response. Clinical decisions affecting the management of ocular infections are based on the identification of the pathogen. When culture results return, the rational question is, "is this bacterium a pathogen or a commensal?" Some species of bacteria are always viewed as pathogens, but many ocular bacteria can be pathogenic or commensal depending on the ocular conditions. That is where the science and art of determining an empirical treatment intersect. Knowledge of normal and pathogenic ocular bacteria is vital for prompt and effective treatment [9, 10]. Objectives of our study was to evaluate the microbiological profile and antibiotic sensitivity pattern of external ocular infection in a tertiary care center.

Material & Methods

The present study was conducted in the Department of Microbiology with the collaboration of Department of Ophthalmology, Patna Medical College & Hospital, Patna, Bihar during a period from August 2023 to November 2023.

Data was collected with irrespective of age and sex. A total of 100 diagnosed cases of external ocular infection patients were enrolled in the present study.

All patients were examined on the slit lamp bio-microscope by the ophthalmologist using standard protocols [11]. After detailed ocular examinations, using standard techniques, specimens for culture and smear were obtained by scraping and swabbing the eyelid margin using sterile blade (# 15) on Bard-Parker handle and sterile broth-moistened cotton swabs in case of blepharitis [12, 13]. Similarly, specimens were also obtained from scraping the corneal ulcers. Conjunctival cultures were obtained by wiping a broth-moistened swab across the lower conjunctival cul-de-sac in conjunctivitis cases. For cases of dacryocystitis purulent material was collected from everted punta by pressure applied over the lacrimal sac area. The obtained ocular specimens were subjected to culture onto the sheep blood agar, chocolate agar, Mac conkey agar, Sabouraud's dextrose agar, thioglycollate medium and brain heart infusion broth. In corneal scraping the specimen was cultured on Blood agar in the form of 'c' shape streak. Direct microscopic examinations such as 10% Potassium Hydroxide (KOH) wet mounting, Gram-stain, Kinyoun's acid-fast stain were also done. For fungus identification Slide culture method and LPCB staining were used.

Microbial cultures were considered significant if growth of the same organism was demonstrated on more than one solid phase medium, and/or if there was a confluent growth at the site of inoculation on one solid medium, and/or if growth of one medium to be consistent with direct microscopy findings (that is, appropriate staining and morphology with Gram stain) and/or if the same organism was grown from repeated specimens [13]. The isolated bacterial strains were identified up to species level by using standard biochemical tests [14]. The antibiotic susceptibility testing was done by the Kirby Bauer disc diffusion method, as per the CLSI guidelines, 2011 [15]. The antimicrobial discs which were used were those of Ampicillin (20µg), Gentamicin (10µg), Amikacin (30µg), Cefazolin (30 µg), Cefuroxime (30µg) Ceftazidime (30µg), Cefotaxime (30µg), Piperacillin/tazobactam(100/10µg), Imipenem (10µg) and Meropenem (10 µg), for the gram-negative bacilli. Penicillin, Ampicillin, Cefoxitin (30µg), Cefotaxime (30µg), Chloramphenicol (30µg), Clindamycin (2µg), Erythromycin (15µg), Oxacillin (1µg), Vancomycin (30µg), Teicoplanin (30µg)), Ciprofloxacin (5µg), Linezolid (30µg) and Tetracycline (30µg) were used to study the susceptibility patterns of the Gram positive cocci . Antibacterial discs were obtained from Hi-Media.

Statistical Analysis

Data was analysed by using simple statistical methods with the help of MS-office software. All the data was tabulated and percentages were calculated.

Observations & Results

In the present study, 100 patients of external ocular infections were enrolled. Among them, 59% were males and 41% were females. Most of the patients (45%) were in age group of >60 years. 16% patients were in age group of <15 years.

Table 1: Age and gender wise distribution of external ocular infection patients.

Age group (years)	Male	Female	Total
< 15	7	9	16(16%)
16-30	8	3	11(11%)

31-45	9	6	15(15%)
46-60	10	3	13(13%)
>60	25	20	45(45%)
Total	59(59%)	41(41%)	100(100%)

In the present study, out of 100 patients, total conjunctival infection was seen in 44(86.27%) patients. Among them, 37(72.55%) cases were conjunctivitis, 6(11.76%) cases were blepharitis and 2(3.92% cases were dacryocystitis. Out of 51 culture cases, keratitis was seen in 7(13.73%) patients. Gram positive cocci was found in 36 patients. And gram-negative bacilli were found in 15 patients on culture. Out of 51 culture, gram positive cocci were 20(39.22%) coagulase negative Staphylococci (CONS) followed by 12(23.53%) Staphylococcus aureus. Gram negative bacilli were (15.69%) Pseudomonas aeruginosa followed by 3(5.88%) Acinetobacter and 2(3.92%) Klebsiella.

Table 2: Showing the bacteria isolated from external ocular infection.

	Organisms	Conjunctival Infection			Total Conjunctival infection	Keratitis	Total
		Conjunctivitis	Blepharitis	Dacryocystitis			
GPC (N=36)	Coagulase negative Staphylococci (CONS)	17	2	0	19(43.18%)	1	20(39.22%)
	Staphylococcus aureus	8	1	1	10(22.73%)	2	12(23.53%)
	Streptococcus pneumonia	2	1	0	3(6.82%)	1	4(7.84%)
GNB (N=15)	Pseudomonas aeruginosa	5	1	1	7(15.91%)	1	8(15.69%)
	Acinetobacter	1	1	0	2(4.54%)	1	3(5.88%)
	Klebsiella	1	0	0	1(2.27%)	1	2(3.92%)
	Citrobacter	1	0	0	1(2.27%)	0	1(1.96%)
	Enterobacter	1	0	0	1(2.27%)	0	1(1.96%)
	Total	37(72.55%)	6(11.76%)	2(3.92%	44(86.27%)	7(13.73%)	51(100%)

In the present study, among keratitis patients, 14 funguses were isolated. Among them, fusarium sps 7(50%), Aspergillus flavus 3(21.43%), Aspergillus niger 2(14.23%), Aspergillus fumigates 1(7.14%) and Candida albicans 1(7.14%) were cultured.

Thus, total number of organisms isolated from external ocular infections was 65, out of which bacterial isolate were 51(78.46%) and fungal isolates were 14(21.54%).

Table 3: Showing fungal isolated in keratitis

Fungus in Keratitis	Number	Percentage
Fusarium species	7	50%
Aspergillus flavus	3	21.43%
Aspergillus niger	2	14.23%
Aspergillus fumigates	1	7.14%
Candida albicans	1	7.14%
Total	14	100%

In the present study, Gram positive isolates were susceptible to vancomycin 100%, teicoplanin 100%, linezolid 100%, clindamycin 86.11%, and ciprofloxacin 83.33%.

Table 4: Antibiotic sensitivity pattern of gram-positive cocci

Antibiotic	No. of patients(N=36)	Percentage
Cefoxitin	25	69.44%
Cefazolin	26	72.22%
Penicillin	29	80.55%
Vancomycin	36	100%
Teicoplanin	36	100%

Netilmicin	33	91.67%
Tetracycline	28	77.78%
Ciprofloxacin	30	83.33%
Clindamycin	31	86.11%
Chloramphenicol	27	75%
Cotrimoxazole	28	77.78%
Linezolid	36	100%
Erythromycin	29	80.56%
Gentamicin	29	80.56%

The Gram negative organisms were mostly sensitive to amikacin 100%, imipenam 100%, meropenam 100% and ciprofloxacin 93.33%, ofloxacin 93.33%, gentamicin 93.33%.

Table.5. Antibiotic sensitivity pattern of gram-negative bacilli

Antibiotics	No. of patients (N=15)	Percentage
Ofloxacin	14	93.33%
Ciprofloxacin	14	93.33%
Gentamicin	14	93.33%
Amikacin	15	100%
Carbencillin	6	40%
Meropenam	15	100%
Imipenem	15	100%
Cefuroxime	10	66.67%
Pip-Taz	13	86.67%
Ampicillin	13	86.67%
Cefazolin	7	46.67%

Discussions

During chronicity of external ocular infection, the disease can affect not only the conjunctiva but also adjacent structures including the eye lid and can be a potential risk for other extra or intraocular infections. Bacteria contribute for about 50–70% of infectious conjunctivitis [16]. Bacterial conjunctivitis is commonly seen in children and the elders but can also be presented among neonates and adults [17,1 8]. Blepharitis which is an inflammation of the eyelid can cause loss of eye lash [19]. The infection may not remain localized and is known to spread to other anatomical sites of the eye [20]. Keratitis, the most serious eye infection is the leading cause of corneal blindness. Moreover, the disease can also progress to endophthalmitis if not diagnosed early [21. 22].

In the present study external ocular infections were predominantly seen in male sex due to their outdoor activities, patients of low socio-economic group [23] like farmers and patients above 60 years of age 45(4%). The study conducted by Srinivasan M et al at Madurai observed patients of low socioeconomic group [23] like farmers were more affected by external ocular infections. The study conducted by Rahman et al., [24] showed that 44.4% of patients belonged to the age group of > 60 years.

Exogenous endophthalmitis is an infective complication of primary cataract, intraocular surgery and ocular trauma due to the introduction of infectious pathogens like bacteria whereas the endogenous one is commonly due to systemic dissemination of the pathogens. Both keratitis and endophthalmitis are potentially devastating ocular infections if not diagnosed early [25, 26]. Dacryocystitis is an inflammation of the nasolacrimal duct. During chronicity the disease is associated with infection, inflammation of the conjunctiva, accumulation of fluid and chronic tearing. This can be potentially dangerous to ocular tissues such as the cornea; leading to post surgery endophthalmitis [27, 28]. Ocular infections, if left untreated, can damage the structures of the eye leading to visual impairments and blindness. Even though the eye is hard and protected by the continuous flow of tear which contains antibacterial compounds, inflammation and scarring once occurred may not be easily resolved and requires immediate management [20].

As seen in Idu F et al., [29] studies Bacterial conjunctivitis was the most commonly seen external ocular infection which was similar in the present study also. The predominant bacterial isolate isolated was Coagulase negative Staphylococci 20(39.22%) which was the commensal of the normal conjunctival flora [30]. The causes of bacterial conjunctivitis were due to the alteration in the normal flora, which can occur by external contamination, by infection spread from adjacent sites or via blood-born path way and disruption of epithelial layer covering the conjunctiva [31].

Staphylococci are associated with any type of eye infections including conjunctivitis, blepharitis, endophthalmitis, keratitis, dacryocystitis and orbital cellulitis; most importantly with blepharitis, conjunctivitis and keratitis [20, 32]. Both S. aureus (Staphylococcus aureus) and CoNS (Coagulase-negative Staphylococci) took the highest proportion of the isolates [33, 34]. Despite their normal existence, CoNS are the most frequent cause of ocular infections with increasing frequencies over time [35]. A 5 year retrospective study in Iran indicated that 40% of infections were due to CoNS [36].

A Similar study in India also found a prevalence of 45.4% [37]. The problem is worse especially in preoperative and post-operative cases. In a study conducted in patients with cataract surgery, 88.8% of isolates from conjunctival swabs were CoNS [38]. Likewise, 65.9% and 21% of pre-operative cataract patients had CoNS and S. aureus isolates respectively. Considering the specific species, S. epidermidis and S. saprophyticus were the common species of CoNS [39]; both species being dominant in subjects with post-operative endophthalmitis as to the study conducted over 20 years in China [40].

In the present study, Gram positive isolates were susceptible to Vancomycin 100%, Teicoplanin 100%, Linezolid 100%, Clindamycin 86.11% and Ciprofloxacin 83.33%. And the Gram negative organisms were mostly sensitive to Amikacin 100%, Imipenam 100%, Meropenam 100% and ciprofloxacin 93.33%, ofloxacin 93.33%, gentamicin 93.33%.

Similar to the study conducted in Tirunelveli – South India [20] where Vancomycin 100% susceptible but Ciprofloxacin is 90% susceptible, the other study conducted in Hyderabad showed Ciprofloxacin 70% sensitive [41]. Resistance and sensitivity based on in-vitro testing may not reflect the true clinical resistance and response to an antibiotic because of the host factors and penetration of the drug [42]. Vancomycin revealed a highest efficacy against Gram positive cocci isolates compared with other antibacterial agents. Vancomycin is a glycopeptide; it inhibits early stages in the cell wall mucopeptide synthesis and it exhibits greatest potency against Gram positive Ocular isolates [42]. Corneal injury was the major cause of corneal ulcer as seen in the study conducted by Chander J Sharma A [43] and Fungi were identified as the predominant aetiological agent for corneal ulceration as in study of Sundaram BM et al., [44]. Both these conditions correlate with the present study. As in the study Fusarium species and Aspergillus flavus were the commonest organism in corneal ulcers which was similar to the study conducted by Venugopal PL-North India [45].

In general, Staphylococcal infection is common in both post infection and post-operative endophthalmitis cases [46, 47]. Moreover, S. aureus and S. epidermidis are known to be the common cause of early onset bleb-associated endophthalmitis [48]. S. aureus is also the threat of eye infection and has been showing significantly increasing trends over time [49]. Among patients with symptoms of conjunctivitis in Nigeria, it was the leading isolate [33]. Comparable findings were also reported in Ethiopia; S. aureus was isolated from 47.6% of blepharitis, 26.6% of conjunctivitis, and 25% of keratitis cases [32].

Conclusions

The present study concluded that the external ocular infection is predominantly more in old age male population. Conjunctivitis is more common external ocular infection. Most common gram-positive cocci isolates are the Coagulase negative Staphylococci (CONS) and Staphylococcus aureus. Pseudomonas aeruginosa is common gram-negative bacilli isolates. Gram positive isolates are more susceptible to vancomycin, teicoplanin, linezolid, clindamycin and ciprofloxacin. Gram negative organisms are more sensitive to amikacin, imipenam, meropenam, gentamicin and fluoroquinolone (ciprofloxacin and ofloxacin).

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