Bacteriological findings in patients with ocular infection and antibiotic susceptibility patterns of isolated pathogens

Khosravi A D, Mehdinejad M, Heidari M

ABSTRACT

Introduction: Isolation of common pathogens involved in ocular infection, and their in-vitro susceptibility to commonly-used ocular antibiotics, as well as the trends in antibiotic resistance developed by these pathogens, were investigated.

Methods: Corneal scrapings were obtained from 318 hospitalised patients and inoculated directly onto enriched and differential culture media. Subcultures were performed on selective media. The necessary biochemical tests were conducted and the organisms identified using standard procedures. Susceptibility of isolated pathogens to commonly-used ocular antibiotics was examined using standard susceptibility testing.

Results: 70 different organisms were isolated. Gram-positive cocci accounted for 47 (67.2 percent) and gram-negative bacilli for 23 (32.8 percent) bacterial isolates. Coagulase-negative Staphylococci (33 percent) and *Pseudomonas* species (24 percent) were the most commonly-isolated organisms. In susceptibility testing, Gentamycin had coverage against 35 (74.5 percent) of 47 gram-positive cocci and 19 (82.6 percent) of 23 gram-negative bacilli tested. The coverage of Tetracycline, Cephalotin and Ceftriaxon against gram-positive cocci were 61.7, 55 and 53 percent, respectively. All the tested gram-positive cocci showed resistance to Cefotaxime and Penicillin. Ceftriaxon and Tobramycin had coverage against 17 (73.9 percent) and 14 (60.8 percent) of 23 gram-negative bacilli isolates, respectively. The coverage of Vancomycin against coagulase-negative Staphylococci was 100%, but all the isolates of *Staphylococcus aureus* were resistant to Vancomycin.

Conclusion: Susceptibility analysis revealed that antibiotic with the greatest coverage was Gentamycin (77.1 percent of 54 isolates). Gentamycin also had good coverage against gram-positive cocci, which constituted the majority (67.1 percent) of ocular isolates.

Keywords: antibiotic susceptibility, bacteria, ocular infection, susceptibility patterns

Singapore Med J 2007; 48(8):741–743

INTRODUCTION

The eye is a unique organ that is virtually impermeable to most environmental agents. Continuous tear flow, aided by the blink reflex, mechanically washes substances from the ocular surface and prevents the accumulation of microorganisms. In addition, lysozyme, lactoferrin, secretory immunoglobulins, and defensins, which are present at high levels in tears, can specifically reduce bacterial colonisation of the ocular surface.[1,2] However in some circumstances, infectious agents gain access to the posterior segment of the eye following one of three routes: (i) as a consequence of intraocular surgery;[3,4] (ii) following a penetrating injury of the globe;[5] or (iii) from haematogeous spread of bacteria to the eye from a distant anatomical site. Although uncommon, endophthalmitis can also result from keratitis, an infection of the cornea with potential complications.[6] Bacterial keratitis is one of the most threatening ocular infections. *Pseudomonas aeruginosa* and *Staphylococcus (S.) aureus* frequently cause severe keratitis that may lead to progressive destruction of the corneal epithelium and stroma.[7,8] Successful treatment of ocular infection, including bacterial keratitis, requires multiple administrations of antibacterial agents to maintain drug concentration in the corneal tissue high enough and for a sufficient period of time to have a useful antibacterial effect.[9] Besides, in the case that the pathogen is not yet known, the choice of antimicrobial agents is commonly made empirically. Where there is access to microbiology facilities, once the organism has been identified, the effective

Department of Microbiology, School of Medicine, Ahwaz Jondi Shapour University of Medical Sciences, PO Box 189, Ahwaz 61335, Iran

Khosravi AD, PhD
Associate Professor

Mehdinejad M, MSc
Instructor

Central Laboratory, Imam Khomeini Hospital, Ahwaz, Iran

Heidari M, BSc
Medical Technologist

Correspondence to:
Dr Azar Khosravi
Tel: (98) 611 3340388
Fax: (98) 611 3332036
Email: khosraviaz@yahoo.com

Table I. Bacterial strains isolated from ocular infections.

Organism(s)	Total no. of isolated recovered	% of isolates
Coagulase-negative Staphylococci	23	32.9
Pseudomonas spp.	17	24.2
S. aureus	9	12.9
S. pneumoniae	6	8.6
Enteric gram-negative bacilli	6	8.6
Alpha-haemolytic Streptococci	5	7.1
Micrococcus	3	4.3
Beta-haemolytic Streptococci	1	1.4

antimicrobial is chosen according to susceptibility testing. This paper report the commonly-isolated pathogens involved in ocular infection. It also investigates the in-vitro effectiveness of commonly-used ocular antibiotics describing their coverage of bacterial species and the trends in antibiotic resistance developed by common ocular pathogens.

METHODS

The study population consisted of 318 hospitalised patients, who mostly underwent operations at the Imam Khomeini Hospital, Ahwaz, Iran, from October 2005 to March 2006. The patients consisted of 138 females and 180 males, with ages ranging from 44 to 83 years, with a mean of 63.6 years. For sampling, corneal scrapings were obtained in the operation room with a sterile blade, using the standard technique.[10] The scrapings were inoculated directly onto blood agar and Thioglycollate broth, and were immediately transferred to the microbiology department central laboratory of the hospital and were incubated at 37ºC for 24 hours. Subculture on blood agar medium, MacConkey agar and Mannitol salt agar (Hi-Media, Mumbai, India) were performed. Blood agar plates were incubated at 5%–10% CO_2 atmosphere. The cultures were examined after 24 hours of incubation. Colony characteristics were studied and cell morphology were examined microscopically. The necessary biochemical tests were conducted and the organisms were identified using standard procedures.[11] Antibiotic susceptibility was determined for all positive cultures using the Kirby-Bauer disc-diffusion method.[12] Antibiotic discs were used (Padtan Teb, Tehran, Iran) and susceptibility of isolated pathogens to commonly-used ocular antibiotics was examined. The results were reported as sensitive, resistant or intermediate resistant with reference to the standard measurement table.[11] The Statistical Package for Social Sciences (SPSS Inc, Chicago, IL, USA) was used for data analysis.

RESULTS

In total, 70 various organisms were isolated from 70 patients. Gram-positive cocci accounted for 47 (67.2%) of all bacterial isolates and gram-negative bacilli for 23 (32.8%) of all bacterial isolates. The isolated organisms and their relative frequency are given in Table I. Coagulase-negative Staphylococci (33%) and *Pseudomonas* spp. (24%) were the most common isolated organisms. Based on results from susceptibility testing (Table II), Gentamycin had coverage against 35 (74.5%) of 47 gram-positive cocci tested isolates and was the most effective antibiotic against *S. aureus*. The coverage of Gentamycin against gram-negative bacilli was 19 (82.6%) of the total 23 tested isolates. The coverage of this antibiotic for *Pseudomonas* spp. was 13 of 17 isolates (76.4%). The coverage of Tetracycline, Cephalotin and Ceftriaxon against gram-positive cocci tested isolates were 61.7%, 55% and 53%, respectively. All the tested isolates of gram-positive cocci showed resistance to Cloxacillin and Penicillin. Ceftriaxon and Tobramycin had coverage against 17 (73.9%) and 14 (60.8%) of 23 gram-negative bacilli tested isolates, respectively. Tobramycin was the most effective antibiotic against *Pseudomonas* spp. in this study and showed good coverage against other isolated enteric bacteria, i.e. Klebsiella, *Escherichia coli*, Proteus and Enterobacter. Amikacin had excellent coverage against *S. aureus* and coagulase-negative Staphylococci. The coverage of Vancomycin against coagulase-negative Staphylococci was 100%. However, all the isolates of *S. aureus* were resistant to Vancomycin.

DISCUSSION

The organisms that cause ocular infection are generally exogenous and result from entry of organisms into the eye during surgery. Sources of contamination include instruments, and the infusion fluids. In addition, the conjunctival sac itself normally harbours several commensals. Contamination of the multidose topical preparations used preoperatively could also be a source.[13] In present study, 70 out of 318 eye samples were culture-positive, so the contamination rate was estimated as 22%. In similar studies, the contamination rates of 25.5%, 43% and 37.7% have been reported.[3,13,14] Similar to these studies, our study has also documented coagulase-negative *Staphylococcus* spp. as the most common isolated organism. Based on the results from susceptibility analysis of the ocular pathogens isolated, the antibiotic with the greatest coverage was Gentamycin (77.1% of 54 isolates). Gentamycin had good coverage against gram-positive cocci which constituted the majority (67.1%) of

Table II. Susceptibility results of various bacteria isolated from ocular infection to commonly-used ocular antibiotics.

Organisms	No. sensitive (%)								
	GM	AM	TE	SXT	CRO	CF	CTX	TOB	K
Coagulase-negative Staphylococci	16(69)	4 (17)	14(61)	11(48)	12(52)	12(52)	–	–	–
Pseudomonas spp.	13(76)	2 (12)	1(6)	4(23)	11(65)	4(23)	9(53)	14 (82)	4 (23)
S. aureus	9 (100)	1(11)	9(100)	8(89)	8(89)	5(55)	–	–	–
S. pneumoniae	3 (50)	1(17)	3(50)	0	3(50)	4(67)	–	–	–
Alpha-Streptococci	3(60)	0	2(40)	0	2(40)	3(60)	–	–	–
Micrococcus	3(100)	2(67)	2(67)	2(67)	3(100)	0	–	–	–
Proteus	3(100)	0	2(67)	2(67)	3(100)	0	–	–	–

GM: Gentamycin; AM: Ampicillin; TE: Tetracycline; SXT: Cotrimaxazole; CRO: Ceftriaxon; CF: Cephalotin; CTX: Cefotaxime; TOB: Ceftriaxon; K: Kanamycin; –: not used

ocular isolates in this study. Besides, the coverage of this antibiotic against gram-negative organisms was also good.

Some of the antibiotics used in present study showed good coverage against the pathogens isolated, for instance, Amikacin against *S. aureus* and Tobramycin against *Pseudomonas* spp. However, some of the other antibiotics showed no effectiveness against tested organisms. The highest resistance was seen in gram-negative and positive cocci to Penicillin and Cloxacillin (98% of 47 isolates), Oxacillin (91.5%) and Ampicillin (87.2%). So, we do not recommend treatment of bacterial keratitis and other ocular infections on an empirical basis, especially in the centres where ophthalmologists have access to microbiology facilities. An attempt should be made to identify the ocular pathogen and performing susceptibility testing. It should be borne in mind that these are in-vitro results and do not always mirror the clinical response to antibiotics due to a variety of reasons including direct topical delivery, corneal penetration of an antibiotic and host factors.[15] However, these results do provide information that allows a clinician to make an informed decision when choosing an initial regimen for treatment of ocular pathogens. Furthermore, the result of the present study provides susceptibility information for ocular pathogens which are resistant to commonly-used ocular antibiotics.

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