Incidence and risk factors of bacteria causing infectious keratitis

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ABSTRACT

Objectives: To study the incidence and risk factors of the bacteria causing infectious keratitis among patients in Qassim province of Saudi Arabia.

Methods: This is a cross sectional study conducted at the Department of Optometry, College of Applied Medical Sciences, Qassim University, Qassim, Kingdom of Saudi Arabia from December 2010 to May 2011. One hundred patients suspected of keratitis were subjected to clinical examinations. A total of 115 corneal swabs from these cases were collected under aseptic conditions for bacteriological examinations.

Results: Culture of the corneal swabs revealed Pseudomonas aeruginosa (25.2%), Staphylococcus aureus (15.7%), and unclassified bacteria (13.9%). However, 52 swabs of infectious keratitis cases (45.2%) were negative to bacteria. Contact lens wearing (44.4%) was the most common risk factor among the examined patients, followed by corneal trauma (21.7%), ocular surface disease (11.3%), and corneal surgery (7%). No significant correlation was observed between systemic risk factor and clinical presentation.

Conclusion: It could be concluded that infectious keratitis was mostly due to Pseudomonas aeruginosa and Staphylococcus aureus. Therefore, strict measures are recommended to control and treat infectious keratitis to avoid visual complications.

Bacterial keratitis is a sight-threatening disease. A special feature is its rapidity of progression and corneal destruction, which may happen within hours by more virulent bacteria. Bacterial keratitis is an ophthalmic emergency, and requires quick and proper treatment as delay of treatment has a considerable

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impact on the severity and duration of disease. Ocular surface supports a small population of naturally inhabitant bacteria as coagulate negative staphylococci (CNS), which have been found to exist as commensals on the mucosa and lid margins. Several ocular disorders are associated with different Gram positive and Gram negative bacteria, including Staphylococcus aureus (S. aureus), Streptococcus sp., Bacillus subtilis, Rhodococcus sp., Pseudomonas aeruginosa (P. aeruginosa), Haemophilus influenzae, Haemophilus aegyptius, and Klebsiella species. Keratitis is an inflammation of the cornea. Bacterial keratitis is an infectious disease of the cornea caused by different bacteria. Severe keratitis can potentially lead to blindness, and in many cases surgical intervention are needed. Bacterial keratitis spreads commonly, in spite of many advances in diagnosis, management, and availability of potent antibiotics. Common use of contact lenses (CLs), ocular surface diseases, corneal trauma, use of immunosuppressive medications and ocular surgery like corneal graft are different types of factors, which cause bacterial keratitis. Contact lens wearing is the leading cause of keratitis in some developed countries, while trauma and ocular surface disease are the leading causes in other countries. Contact lens wearing is one of the greatest risk factors for infective keratitis, and may account for 20-50% of all cases. Contact lens related bacterial keratitis are usually caused by Pseudomonas species. These bacteria are very virulent and can be visually devastating because of their ability to alter genes, which are related to virulence, survival, and adaptation. The S. aureus, S. pneumoniae, and Pseudomonas species are the common types of bacteria causing corneal ulcers worldwide. P. aeruginosa is the most common and virulent ocular pathogen, and possesses a combination of unique bacterial virulence characteristics, such as increased binding of bacteria to corneal epithelial cells possibly through exposure of specific bacterial adhesins on cell membranes, increased internalization of bacteria through expression of membrane lipid rafts on corneal epithelial cells, and expression of exoenzymes, which is injected into the host cell and initially locate to the plasma cell membrane, invade epithelial cells, replicate intracellularly, and produce cell death through disruption of the host cell actin cytoskeleton and intracellular phospholipase A2 activity. The present study aimed to investigate the incidence and risk factors of infectious keratitis together with isolation and identification of the causative bacteria.

Methods. This was a cross-sectional study conducted at the Department of Optometry, College of Applied Medical Sciences, Qassim University, Qassim, Kingdom of Saudi Arabia from December 2010 to May 2011 in collaboration with King Fahd Specialist Hospital Buraidah, which is a tertiary care hospital in Qassim Province. Patients coming directly to the emergency department or referred from peripheral basic health units, or ophthalmologists were included in this research project. One hundred patients suspected of having bacterial keratitis due to painful red eyes were subjected to detailed clinical examination including visual acuity, corneal epithelial defects, number and position of corneal infiltrates, and anterior chamber reaction.

Sampling. Under aseptic conditions, 115 corneal swabs/scrapings were obtained from 100 patient’s suspected of bacterial keratitis for bacteriological examinations. The samples were transferred immediately to the laboratory for processing.

Bacterial isolation and identification. This technique was carried out routinely in all collected samples including culturing, sub-culturing and purification, isolation, and identification. The collected swabs were inoculated in tryptic soya broth overnight at 37°C. Consequently, the broth was inoculated onto blood agar, MacConkey’s agar, mannitol salt agar, and chocolate agar media, and then incubated aerobically at 37°C for a maximum up to 48 hours. Inoculated chocolate agar plates were left in anaerobic incubator at 5% CO2. All the bacterial isolates were identified by their colony morphology, Gram staining, pigment production, relevant biochemical tests, and API strips according to the manufacturer. The minimum criteria for the positive culture were considered upon the growth of 3 colonies on one solid medium.

Statistical analyses. Data were recorded in Microsoft Access and manipulated with Microsoft Excel. Data were imported into Statistical Package for Social Sciences version 14 (SPSS Inc, Chicago, IL, USA) for data analysis. For purposes of data analysis, patients of the 4 major risk factor groups only were used, namely: CL wear; ocular surface disease; ocular trauma; and ocular surgery. The statistical level of significance was set at 5%.

Results. Patient’s clinical characteristics. A total of 100 patients (115 eyes) with a diagnosis of bacterial keratitis were examined during the study period of 6 months. Out of 100 subjects, 85 cases were examined for the first time in the emergency department of our hospital, whereas 15 cases were referred by general practitioners or ophthalmologists. The age of the patients ranged from 12-55 years (mean age: 21 years). Gender distribution was 23 men and 77 women. Most patients were living in urban areas.
**Frequency of predisposing ocular conditions.** The examined patients revealed that CL wearing was the most common risk factor, and was found in 51 eyes (44.4%), and no risk factors were identified in 18 eyes (15.7%) of cases. Table 1 summarizes all the predisposing factors, and describes that CL-wearing was the most common risk factor among patients of infectious keratitis followed by corneal trauma. Diabetes mellitus was a risk factor in 20 cases, but there was no significant correlation between systemic risk factors and severity of clinical presentation. The clinical course of these patients was acute with a lid and conjunctival edema, reduced vision, pain, redness, severe photophobia, and discharge. Unilateral right eye was observed to be involved by keratitis in (51%) and unilateral left eye in (34%) of patients. Infection was bilateral in 15 patients (15%). Visual acuity at the time of examination ranged from 20/20 to 20/200. Corneal infiltrates were single in 84 eyes (73%), and multiple in 31 (27%). Nasal infiltrates were most common in 47 (40.9%). In addition to this, anterior chamber inflammation was absent in 40.9% (n=47) of cases. A 1+ to 2+ Tyndall effect was present in 57.4% (n=66) of cases, whereas severe anterior chamber inflammation (3+ to 4+), and hypopyon were present in 1 (0.9%) each. The location of the infiltrates was distributed as shown in Table 2.

**Microbiological characteristics.** Culture results of the collected corneal scraping swabs revealed the isolation of bacteria from 63 swabs (54.8%) were *P. aeruginosa* (n=29, 25.2%), *S. aureus* (n=18, 15.7%), and unclassified bacteria (n=16, 13.9%) were isolated (Table 3). However, 52 (45.2%) swabs of infectious keratitis cases were negative to bacteria. Also, multiple organisms were found in 8 (7%) swabs from corneal scraping, and the Gram negative were the predominant type of bacteria. In the CL-wearing group, 26 (51%) of the corneal scrapings were positive, and the cultured bacteria were *P. aeruginosa* in 21 (80.8%) of cases. The percentage of Gram positive bacteria was 5 (38.5%) in the ocular surface disease group, and 8 (32%) in the corneal trauma group. Mixed bacteria not fitted in specific group were also cultured in 16 (13.9%). The *P. aeruginosa* was the most common culture lead to CL-related cases (80.8%, *p*<0.001), and *S. aureus* was most common in ocular surface disease group (38.5%, *p*<0.001). The severity of patients’ keratitis at presentation varied significantly by their culture result. The *P. aeruginosa* had statistically significantly more severe keratitis at the time of scraping as compared with other microorganisms (*p*<0.05).

**Table 1 -** Recorded predisposing risk factors among patients suffering from infectious keratitis included in a study conducted at the Department of Optometry, College of Applied Medical Sciences, Qassim University, Qassim, Kingdom of Saudi Arabia.

<table>
<thead>
<tr>
<th>Factors</th>
<th>Number of swabs (n=115)</th>
<th>Incidence (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Contact lens wear</td>
<td>51</td>
<td>(44.4)</td>
</tr>
<tr>
<td>Corneal trauma</td>
<td>25</td>
<td>(21.7)</td>
</tr>
<tr>
<td>Ocular surface disease</td>
<td>13</td>
<td>(11.3)</td>
</tr>
<tr>
<td>Corneal surgery</td>
<td>08</td>
<td>(7.0)</td>
</tr>
<tr>
<td>None</td>
<td>18</td>
<td>(15.7)</td>
</tr>
</tbody>
</table>

**Table 2 -** Location of the corneal infiltrate among patients suffering from infectious keratitis included in a study conducted at the Department of Optometry, College of Applied Medical Sciences, Qassim University, Qassim, Kingdom of Saudi Arabia.

<table>
<thead>
<tr>
<th>Position of corneal infiltrates</th>
<th>No. of eyes</th>
<th>Incidence (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Temporal</td>
<td>41</td>
<td>(35.7)</td>
</tr>
<tr>
<td>Nasal</td>
<td>47</td>
<td>(40.9)</td>
</tr>
<tr>
<td>Central</td>
<td>17</td>
<td>(14.8)</td>
</tr>
<tr>
<td>Diffuse</td>
<td>10</td>
<td>(8.7)</td>
</tr>
</tbody>
</table>

**Discussion.** In the present study, the incidence of culture positive microbial keratitis was 54.8% among the population of Qassim Province. Among the isolated bacteria, *P. aeruginosa* was the most common cultured bacteria followed by *S. aureus*. Parallel studies in other regions of the world conducted on large scales have shown varying degrees of culture positive bacteria. Pachigolla et al16 showed that the eyes of 131 patients that underwent 139 corneal scrapings was presumed to be microbial keratitis. They also observed that *P. aeruginosa* was isolated from 73 cases (52.5%).16 Passos et al17 in their study showed culture positivity of 53.5% organisms (bacteria - 47%, fungi - 6.1%, and acanthamoeba - 0.4%). The most frequent bacteria were
the Gram-positive cocci (mostly coagulase-negative *Staphylococci*) and Gram-negative bacilli (mostly the genera *Pseudomonas*, *Moraxella*, and *Proteus*). Green et al. showed that cultures of corneal scrapings were positive in 65% of cases, where *P. aeruginosa* (n=44; 17%), coagulase-negative staphylococci (n=22; 9%), *S. aureus* (n=19; 8%), and fungi (n=7; 3%) were commonly recovered.

In our study, *P. aeruginosa* was the most common bacteria cultured (25.2%) and similarly the above mentioned studies also show that this bacteria is the most common cultured followed by *Staphylococci*, although the culture positive rate for pseudomonas is higher as compared to our study. This variation may be due to different risk factors in these studies. A higher rate of *P. aeruginosa* isolation from CL-related bacterial keratitis was also found in some other countries, such as Australia (55%) in 2008, Malaysia (64%), and in Australia in 2007. Tremendous use of CL during the last decade increased the risk factor of microbial keratitis dramatically. This was also observed in our study (44%), it contributed to almost 50% cases of culture positive microbial keratitis for *P. aeruginosa*. Corneal surface diseases and corneal trauma have been found to be second most common cause of bacterial keratitis, accounting for 33% of cases. Willcox also observed that *P. aeruginosa* is usually the most common bacterial pathogen isolated from cases of keratitis. This infection poses a serious threat to normal vision, and is associated with extended wear of CL and eye trauma. Tissue damage during bacterial keratitis results from the action of bacterial products on ocular tissues and from the host inflammatory response to the infection. The *P. aeruginosa* has the ability to attach and grow on lens materials, and survive in CL storage cases because of production of resistant biofilm, and partly due to resistance to CL disinfectants.

Although infectious keratitis is caused by many bacteria, fungi, acanthamoeba and/or viruses, our current study was limited to most prevalent bacteria contributing to such infections (*P. aeruginosa* and *S. aureus*).

In conclusion, as infectious keratitis represents a potentially significant threatening ocular disease, initial diagnosis facilitates its resolution in the hope of preventing future visual loss. In this study, we conclude that 54.8% of microbial keratitis among patients in Qassim Province was caused by bacteria mainly *P. aeruginosa* and *S. aureus*. Therefore, strict measures are recommended to control and treat infectious keratitis.

**References**


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