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Human microbial keratitis in Upper Egypt

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Abstract: One hundred and fifteen cases of keratitis were examined both epidemiologically and microbiologically. Direct microscopic examination and culturing of eye specimens from these cases revealed that keratitis was mostly due to bacteria (50 cases) followed by fungi (30 cases) and mixed fungal and bacterial keratitis (5 cases), while 30 cases were negative for bacteria and fungi. *Staphylococcus aureus* and other *Staphylococcus* species were the most dominant etiologic agents of bacterial keratitis. *Aspergillus flavus, A. terreus, A. niger*, *Candida albicans* and *Cladosporium cladosporioides* were common species associated with fungal keratitis. Adults between the ages of 31-70 years (72.2 % of the 115 total number of cases studied) were most susceptible to keratitis as well as in the case for mixed fungal and bacterial keratitis. Males predominated (71.3%) while females made up 28.7% of total number of cases. Many males were farmers, i.e. (40% of the total number of cases). Corneal trauma proved to be the most prevalent risk factor of keratitis and occurred in 63.4 % of the total number of cases). Trauma was reported in 78 % of cases in which 49.6% of these cases were from plant origin. To our knowledge, *F. proliferatum, Trichoderma hamatum* and *Stemphylium botryosum*; are recorded for the first time from cases of keratitis.

Key words: corneal ulcer, bacterial keratitis, mycotic keratitis, trauma, cataract surgery.

Introduction

Corneal ulcers are due to exogenous infections, with the organisms entering through the corneal epithelium (Doczi *et al*. 2004). Scarring of the cornea as a result of suppurative keratitis, is an important cause of preventable blindness. In some developing countries, corneal infections are the second commonest cause of blindness after unoperated cataract (Whitcher *et al.* 2001). Suppurative corneal ulcers may be caused by bacteria, fungi, and protozoa. However, in the tropics, up to two thirds of ulcers may be due to filamentous fungi. This type of ulceration is commonly associated with ocular trauma (Srinivasan *et al*. 1997, Khanal *et al*. 2001). Specific treatment requires prompt and accurate identification of the causative microorganisms (Foster 1992).

Bacterial keratitis, because of its high incidence and potential complications, is one of the most threatening ocular infectious pathologies that can lead to severe visual disability (McLeod *et al*. 1995) or corneal perforations (Ostler 1993). The severity of the corneal infection usually depends on the underlying condition of the cornea and the pathogenicity of the infecting bacteria (Vajpayee *et al*. 2000).

Bacterial keratitis is rare in the absence of predisposing factors (Benson & Lanier 1998). Until recently, most cases of bacterial keratitis were associated with ocular trauma or ocular surface diseases. However, the widespread use of contact lenses has dramatically increased the incidence of contact lens-related keratitis (Schaefer *et al*. 2001).

The spectrum of bacterial keratitis can also be influenced by geographic and climatic factors. Many differences in keratitis profile have been noted between populations living in rural or in city areas, in western, or in developing countries (Schaefer *et al*. 2001). In addition, emergence of multiresistant bacterial strains is a major concern when antibiotics such as fluoroquinolones are used as monotherapeutic agents (Schaefer *et al*. 2001).

Species of *Aeromonas*, *Bacillus*, *Citrobacter*, *Enterobacter*, *Escherichia*, *Haemophilus*, *Klebsiella*, *Micrococcus*, *Mycobacterium*, *Proteus*, *Pseudomonas*, *Serratia*, *Staphylococcus* and *Streptococcus* were isolated from corneal ulcers (Srinivasan *et al*. 1997, Alghalibi 2000, Leck *et al*. 2002, Fong *et al*. 2004)). *Escherichia coli* was also found to cause bacterial keratitis in people wearing contact lenses (Houang *et al*. 2001).

Fungal keratitis may account for more than 50% of all cases of culture-proven microbial keratitis and of ophthalmic mycoses, especially in tropical and subtropical areas (Hagan *et al*. 1995). Filamentous fungi are the principal cause of mycotic keratitis in most parts of the world. *Aspergillus* (*A. flavus*, *A. fumigatus*, *A. nidulellus*, *A. niger*, *A. terreus*) (Dunlop *et al*. 1994, Khairallah *et al*. 1992) or *Fusarium* (*F. dimerum*, *F. oxysporum*, *F. solani*, *F. verticillioides*) (Rosa *et al*. 1994, Hagan *et al*. 1995, Srinivasan *et al.* 1997, Wong *et al*. 1997) are the most common causes. Moreover, *Acremonium* (*A. kiliense*, *A. potronii*), *Bipolaris* (*B. hawaiiensis*, *B. spicifera*), *Cephaliophora irregularis*, *Cladosporium cladosporioides*, *Curvularia* (*C. geniculata*, *C. lunata*, *C. senegalensis*), *Exserohilum rostratum*, *Paecilomyces* (*P. lilacinus*, *P. variotii*), *Penicillium* spp. and *Scedosporium apiospermum* are the other most important causes of keratitis in a number of studies (Rosa *et al*. 1994, Srinivasan *et al*. 1997, Garg *et al*. 2000, Leck *et al*. 2002, Doczi *et al*. 2004), while the coelomycete *Lasiodiplodia theobromae* has been reported to cause keratitis in India (Thomas 1990, Thomas *et al*. 1998, Garg *et al*.

2000) and the southern U. S. (Liesegang and Forster 1980, Rosa *et al*. 1994). In addition, Al-Hussaini (1990) and Alghalibi (2000) reported the following species but less commonly: *Alternaria alternata*, *A. chlamydospora*, *Aspergillus ochraceus*, *A. sydowii*, *A. ustus*, *A.versicolor*, *Candida albicans*, *Candida* spp., *Cladosporium herbarum*, *Conidiobolus coronatus*, *Derchslera spicifera*, *Mucor hiemalis*, *Oidiodendron rhodogenum*, *Penicillium chrysogenum*, *P. citrinum*, *P. funiculosum*, *Rhizopus stolonifer*, *Trichothecium roseum* and *Ulocladium chartarum*.

Most of the studies done exclusively on mycotic keratitis have listed trauma as being the most common risk factor (occurring in 44 to 55% of patients). Other factors include prolonged use of topical corticosteroids or antibacterial drugs, systemic diseases such as diabetes mellitus, preexisting ocular diseases, and wearing contact lenses (Al-Hussaini 1990, Rosa *et al*. 1994, Panda *et al*. 1997, Alghalibi 2000, Tanure *et al*. 2000, Xie *et al*. 2001, Gopinathan *et al*. 2002). Some studies have sought to compare the most frequent risk factors in mycotic and bacterial keratitis (Dunlop *et al*. 1994, Wong *et al*. 1997). Interestingly, topical corticosteroid therapy, which is frequently perceived to be a specific risk factor for mycotic keratitis, appeared to predispose less frequently to mycotic keratitis (25%) than to bacterial keratitis (38%) (Wong *et al*. 1997).

Filamentous fungal keratitis appears to occur most commonly in healthy young men engaged in agricultural work or outdoor occupations. Various traumatizing agents have been reported, including vegetable matter, mud or dust particles, paddy grain, the swish of a cow's tail, tree branches, and metallic foreign bodies (Rosa *et al*. 1994, Gopinathan *et al*. 2002).

Keratitis due to yeasts and yeast-like fungi is most frequently caused by *C. albicans* (Liesegang & Forster 1980, Forster 1994, Tanure *et al*. 2000). However, *C. albicans* and related fungi have been infrequent isolates in more recent studies performed in tropical countries (Dunlop *et al*. 1994, Hagan *et al*. 1995, Srinivasan *et al*. 1997, Gopinathan *et al*. 2002, Leck *et al*. 2002), possibly due to the predominance of livelihoods, such as agriculture, which carry a higher risk for the occurrence of trauma-related keratitis caused by filamentous fungi than for keratitis due to *C. albicans*.

This study was designed to investigate cases of human keratitis in Upper Egypt. Sex, age, occupation and predisposing factors were also considered.

Materials and Methods

1. Clinical diagnosis and sampling of keratitis specimens

Patients

A total of 115 patients clinically diagnosed to have microbial keratitis were investigated. The patients were admitted and treated in the Department of Ophthalmology of Assiut University Hospitals during the period from January, 2005 to April, 2006. All patients were from Upper Egypt. Each patient was assigned a case number. Date of sampling, sex, age, occupation and place of residence were recorded from the regular hospital records. A history of the circumstances of how the eye became infected and possible predisposing factors was recorded.

Specimens collection

Corneal scrapings were performed under aseptic conditions by an ophthalmologist using a sterile surgical blade No. 15. Scraping was performed at a slit-lamp biomicroscope after instilling topical benoxinate hydrochloride 0.4% (Thilo) into the eye to ensure that adequate material was obtained and to avoid perforation of the eye (Srinivasan *et al.* 1997). Occasionally: ulcers were very soft, sticky and had a mucoid consistency in which case they were too slippery and tenacious to be removed with a metal scraper. A disposable sterile cotton swabs was used (Jacob *et al*. 1995).

2. Direct microscopic examination and culturing of keratitis specimens

Corneal scrapings or swabs were subjected to both direct microscopic examination and culturing.

Direct microscopic examination

A potassium hydroxide (10%) wet mount was used in examining the direct smears to identify cells, hyphae or other fungal elements (Arffa *et al*. 1985). Lactophenol cotton blue (Kwon-Chung & Bennett 1992) and Gram stain (Cheesbrough 1992) were also used. Gram stain allows for the detection of both fungi and bacteria. However, it does not stain the cell wall or septa of fungal hyphae, but is absorbed by the protoplasm. Most bacteria can be differentiated by their Gram reaction.

Culturing of bacteria and fungi

The material collected by scraping or swabbing was inoculated directly into four agar medium plates. Blood and Endo agar (Ronald 2000) were used to identify bacteria, while Sabouraud dextrose agar (Moss & McQuown 1969) and Czapek's glucose agar (Smith & Dawson 1944) were used to identify fungi. Due to the small amount of specimen that can be collected in cases of keratitis, only one plate of each medium was used.

Inoculation procedure

All agar plates were inoculated by lightly streaking both sides of the surgical blade or cotton swab applicator over the agar surface in C-streaks, taking care not to penetrate the agar surface (Benson

& Lanier 1992). After inoculation the bacterial plates were incubated at 37°C for 24-48 hrs then left for a week to detect slow growing bacteria, while the plates of Sabouraud dextrose agar and Czapek agar were incubated at 28° C for a week in order to isolate fungi. Growth on the C-streaks is considered to be significant, while growth outside the C-streaks is likely to be caused by contamination.

3. Identification of bacteria and fungi

Bacteria were identified by standard microbiological methods. Culture characters of pure colonies, bacterial morphology using Gram stain, hemolysis on blood agar, biochemical tests including the coagulase test for *Staphylococcus* species, optochin sensitivity for *Streptococcus* species, Cytochrome C oxidase, pigment production and growth at 42°C for *Pseudomonas* species, lactose fermentation, indol test, methyl red test, Voges-Proskauer's test, citrate test, urea utilization and growth on Endo agar for Enterobacteriaceae were tested. The following references were used for identification of bacterial genera and species: Stolp & Gadkari (1981), Holt *et al.* (1994), Brooks *et al*. (1998) and Champoux *et al*. (2004).

For identification of fungal genera and species (based on macro-and microscopic characteristics), the following references were used Raper and Fennell (1965), Domsch *et al*. (1980), Moubasher (1993), De Hoog *et al*. (2000), Leslie and Summerrell (2006). Chromagar was also used for confirmation of *Candida* isolates.

Results and Discussion

Microbial keratitis cases studied

Of the 115 cases of keratitis studied and as determined by either or both culturing and direct microscopic examination methods, 50 were due to bacterial infection, 30 were due to fungal infection and 5 cases were due to mixed bacterial and fungal infections, while the other 30 cases were negative for both bacteria and fungi (Table 1). The results reveal that keratitis cases due to bacterial infections were more common than those due to fungal infections. This finding correlates well with what is previously reported in the literature (Alghalibi 2000, Fong *et al*. 2004 and Keay *et al*. 2006). However, Srinivasan *et al*. (1997) in their study of 434 cases, found that 140 cases were due to pure bacterial infection, 139 to pure fungal infection and 15 due to mixed bacterial and fungal infection while the rest of the cases (137) were negative for both bacteria and fungi.

In the current study, seven bacterial species were isolated from the 50 pure bacterial keratitis cases, all of which were isolated on blood agar medium while only one species was cultured on endo agar medium. From these results it can be concluded that blood agar is the most suitable medium for the isolation of bacteria from keratitis cases. Our results are supported by the results of Srinivasan *et al*. (1997), Alexandrakis *et al*. (2000), Houang *et al*. (2001), Leck *et al*. (2002), Kaye *et al*. (2003), Fong *et al*. (2004) and Keay *et al*. (2006).

Forty nine bacterial cases were due to only one bacterial species, while only one case was due to two different bacterial species. Keratitis cases due to one bacterial species have been commonly reported by Schaefer *et al*. (2001), Leck *et al*. (2002) and Fong *et al.* (2004). However, a few cases were reported to be due to two species (Hall & McKellar 2004) or more (Bourcier *et al*. 2003, Butler *et al*. 2005, Keay *et al*. 2006).

Species of *Staphylococcus* were the most common agents in 45 cases out of 50. Earlier reports are in agreement with our finding (Hall & McKellar 2004, Butler *et al*. 2005, Keay *et al*. 2006). *S. aureus* was the most common species involved in 30 out of the 50 cases. This result is in agreement with Alghalibi (2000) who reported 31 cases as due to *S. aureus* out of 94 bacterial cases. An unidentified species of *Staphylococcus* was isolated from 15 cases (30%). Similarly, unidentified *Staphylococcus* spp. were reported from 19.4% (Hall & McKellar 2004), 27.8% (Butler *et al*. 2005), and 16% of the cases (Keay *et al*. 2006).

Pseudomonas aeruginosa was isolated from two cases out of the 50. *P. aeruginosa* was the most common agent related to wearing contact lenses, representing 13% of the cases investigated in Hong-Kong (Houang *et al.* 2001) and 37.3% in Taiwan (Fong *et al*. 2004). However, in the present study *Staphylococcus aureus* was the etiological agent isolated from only one case out of two cases examined where contact lenses were worn. The other four bacterial species (*Escherichia coli*, *Klebsiella pneumoniae*, *Micrococcus luteus* and an unidentified actinomycete) were isolated each from only one case (Table 1). *Actinomyces* species, *Klebsiella pneumoniae* (Bourcier *et al*. 2003), *E. coli* (Houang *et al.* 2001) and *Micrococcus* species (Leck *et al*. 2002) have also been reported in few cases before.

Direct microscopic examination of 44 bacterial cases revealed that 38 cases were positive with Gram stain while all were negative with 10% KOH and LPCB. This is in agreement with the findings of Fong *et al.* (2004) that Gram stain gave positive results in 72.5% out of 229 smears studied. Our results revealed that Gram stain is the most suitable method for direct microscopic examination of bacterial keratitis specimens. However, the 50 cases when tested by culturing method, were all found to be positive for bacterial growth. Fong *et al*. (2004) in their study of microbial keratitis in a University Hospital in Taiwan using the two microbiological methods found that most cases were positive by culturing and/or direct microscopic examination;

however some cases were negative by culturing and positive by smears and vice versa.

Twelve fungal species were isolated from the 30 fungal keratitis cases on Sabouraud's agar. Only six specimens showed fungal growth on Czapek's agar. Fungal growth was observed in 26 out of these 30 cases on blood agar. This implies that Sabouraud's medium was the most suitable to culture fungi from eye specimens. Other studies reported similar findings (Garg *et al*. 2000, Houang *et al.* 2001, Leck *et al*. 2002, Marangon *et al*. 2004, Butler *et al*. 2005 and Pate *et al*. 2006).

Only one fungal species was recovered from each of the 30 fungal keratitis cases. Similar results were found by Garg *et al*. (2000), Leck *et al*. (2002) and Marangon *et al*. (2004).

Aspergillus species were the most common fungi, involved in 18 out of the 30 fungal cases (Table 1). In agreement with our results, *Aspergillus* spp. were reported from 48.5% (Al-Hussaini 1990), 37% (Dunlop *et al*. 1994), 39.5% (Panda *et al*. 1997) and 65.7% of total fungal cases (Alghalibi 2000). *Aspergillus flavus* was the most common species recorded from 10 out of the 18 cases. *A. terreus* came second (4 cases) followed by *A. niger* (3 cases) and *A. fumigatus* (1 case) (Table 1). In Upper Egypt, Al-Hussaini (1990) recorded *A. fumigatus* from 12 cases, *A. niger* (6) and *A. flavus* (5) out of 67 fungal keratitis cases. Ten years later and from patients of the same area, Alghalibi (2000) isolated *A. flavus* from 10 cases, followed by *A. fumigatus* and *A. niger* (9 cases each), *A. ochraceus* and *A. versicolor* (2 each), *A. sydowii* and *A. ustus* (1 each) out of 70 fungal cases. Leck *et al*. (2002) reported *A. flavus* from 59 cases in India and 9 cases in Ghana, *A. fumigatus* from 15 cases in India and 7 in Ghana, *A. niger* from one case in each country out of 353 and 109 fungal keratitis cases in India and Ghana respectively. In USA, Marangon *et al*. (2004) isolated *A. fumigatus* from 9 cases, *A. flavus* and *A. terreus* from 3 cases each and *A. niger* from 2 cases out of 421 fungal keratitis cases.

Candida was reported from 3 cases (represented by *C. albicans* from 2 cases and *C. krusei* from one) (Table 1). Leck *et al*. (2002) and Marangon *et al*. (2004) reported *Candida* species each from 1 case (0.9% and 0.24%, respectively) of total cases they studied.

Cladosporium cladosporioides was recovered from two cases only. Leck *et al.* (2002) also isolated this fungus only from 1 case (0.9%) of 109 fungal cases studied in Ghana. In Upper Egypt, Al-Hussaini (1990) and Alghalibi (2000) isolated *C. herbarum* from 22.7% and 2.9% of total keratitis cases they studied, respectively.

Cochliobolus spicifer, three *Fusarium* species (*F. oxysporum*, *F. proliferatum* and *F. solani*), *Penicillium chrysogenum*, *Trichoderma hamatum* and sterile mycelia were reported each from one case of the 30 fungal keratitis cases studied (Table 1). *Cochliobolus spicifer*, *F. oxysporum*, *F. solani* and *Penicillium chrysogenum* were also isolated from a few cases by Al- Hussaini (1990) and Alghalibi (2000). However, species of *Fusarium* were reported to be the most predominant etiological agents of keratomycosis in surveys of Srinivasan *et al*. (1997), Wong *et al*. (1997), Leck *et al.* (2002) and Fong *et al*. (2004), where they were reported from 43%, 51.7%, 41% and 10% of the total fungal keratitis cases studied respectively.

Direct microscopic examination and culturing methods of the 30 fungal cases, revealed that all the specimens gave fungal growth, while out of 17 cases tested by 10% KOH, LPCB, and/or Gram stain, only 8 showed fungal elements and 9 showed no elements at all. Out of these 17 fungal cases tested using 10% potassium hydroxide solution and lactophenol cotton blue, 6 and 5 cases showed fungal elements, respectively, while only two out of 16 specimens using the Gram stain, showed fungal elements. It is therefore concluded that 10% potassium hydroxide solution and lactophenol cotton blue are the most suitable methods for direct microscopic examination of fungal cases. Similarly Alghalibi (2000) found that 10% KOH and LPCB showed fungal elements in 40 and 36 fungal cases respectively, out of 70 cases tested.

In the current study, of 115 keratitis cases investigated, only 5 cases were caused by a mixture of bacterial and fungal agents. Four of these mixed cases were due to only one bacterial and one fungal species while the fifth case was due two different bacterial and one fungal species. Srinivasan *et al*. (1997) reported 15 (3.4%) mixed microbial keratitis cases out of 434 keratitis cases studied in India, each infected by a single species of bacteria mixed with a single species of fungi. Leck *et al*. (2002) reported also 48 (6.9%) mixed cases by one bacterial and one fungal species out of 1090 keratitis cases studied in Ghana and India.

Staphylococcus aureus was recovered from four polymicrobial cases while *Klebsiella pneumoniae* and an unidentified *Staphylococcus* species were recovered each from one case (Table 1). *Aspergilus flavus* was isolated from two cases associated with a species of *Staphylococcus*, while *Fusarium verticillioides* and *Stemphylium botryosum* were isolated each from one case and found asscociated with *S. aureus*. On the other hand *Cladosporium cladosporioides* was isolated from one case and found mixed with two bacterial species (*S. aureus* and *K. pneumonia*). *Staphylococcus* species were also relatively prevalent in mixed keratitis cases studied by Pate *et al*. (2006) in USA where they were isolated from 36% out of 89 bacterial isolates.

From the available literature, it is worthmentioning that *Fusarium proliferatum*, *Trichoderma hamatum* (from pure fungal keratitis cases) and *Stemphylium botryosum* (from mixed keratitis case) were not reported earlier.

The four mixed cases tested by direct microscopic examination showed fungal elements and/or bacterial cells (using 10% KOH, LPCB and Gram stain). Pate *et al*. (2006) examined 52 corneal smears using Gram stain and found that 10 showed only fungal elements, 10 showed fungi and bacteria while 16 showed only bacterial cells.

Mixed infection may be due to differences in risk factors, climates and access to care. This polymicrobial synergism can influence the severity of an infection (Pate *et al*. 2006).

Incidence of keratitis in relation to age and sex of patients

The most susceptible persons to keratitis were adults of age 31-70 years (72.2% of 115 cases studied) with the highest number of cases being recorded among patients of 51-60 years (20.9% of the total cases) (Table 2). Generally keratitis cases due to bacteria were greater than those caused by fungi and mixed cases. The most susceptible persons for bacterial keratitis were those of age 41-50 years followed by those of age 61-70 years while in case of fungal keratitis the most susceptible persons were those of age 51-60 years. So, it could be concluded that the susceptibility of adults (over 50 year old) for keratitis may be due to immunity factor.

Keratitis in general showed also a correlation with sex of patients where males were more often contracted the disease than females (71.3% versus 28.7% of 115 cases investigated). This pattern was seen in keratitis cases caused by bacterial or fungal agents. In accordance with our results, Srinivasan *et al*. (1997) found that the most susceptible persons to keratitis were adults of 31-60 years (59% of 434 cases studied) with males being more often affected than females (ratio of 2:1). Garg *et al.* (2000) found that the average age in keratitis was 46.5 years with male to female ratio of 4:1. Butler *et al.* (2005) found 103 (54%) patients were males and 87 (46%) were females out of 190 patients studied for keratitis with a mean of age 75.5 years (range 60-101 years).

Incidence of keratitis in relation to occupation of patients

Most patients with keratitis (out of 115 cases studied) were farmers (40% of the cases) followed by housewives (16.5%, most of them live in rural areas) (Table 3). This pattern was seen in cases caused by bacterial or fungal agents. It is obvious that in the cases of males, occupational risk plays a big role in contracting the disease. The same result was seen in a study in India where the majority of 434 kerattis cases were farmers (56.4%) followed by housewives (12% of total cases) (Srinivasan *et al*. 1997). Similar findings were also reported in Egypt (Alghalibi 2000) and China (Xie *et al*. 2001). Other occupations (e.g. workers, retirees, students, children, drivers, employees and soldiers) were represented by lower percentages of keratitis cases (0.9–14.8%). These results are in agreement with those reported by Srinivasan *et al*. (1997) and Alghalibi (2000).

Incidence of keratitis in relation to risk factors

Trauma of the corneal tissue of eye was the most significant risk factor of keratitis where 63.4% of the 115 cases were due to trauma. Most cases of trauma originated from contact with plants (78% of trauma cases). This pattern was seen in cases of keratitis caused by bacteria, fungi or both (Table 4). Foster *et al*. (1981) reported trauma in 736 (54.4%) out of 1352 keratitis patients of which 188 cases were due to contact with plant material. Srinivasan *et al*. (1997) found also that the most predisposing factor for keratitis was corneal trauma in 284 (65.4%) out of 434 cases. They also reported that trauma due to paddy leaf was represented by 25.4% of the total number of cases; tree branch or thorn by 18.7%; dust, soil or stones by 18%; vegetable matter by 15.1%; animal matter by 5.6%; metallic foreign body by 4.9%; fingernail by 4.2% and other unknown factors by 8.1% of 284 trauma cases. Keay *et al*. (2006) also reported trauma as the most common risk factor in 106 (36.4 %) out of 291 keratitis cases studied.

Risk factors other than trauma, e.g. cataract surgery, diabetes, hypertension, wearing contact lenses, immunosuppression and other unknown factors, showed lower percentages (Table 4). Similar findings were reported by Foster *et al*. 1981, Al-Hussaini 1990, Rosa *et al*. 1994, Panda *et al*. 1997, Wong *et al.* 1997, Yee *et al*. 1997, Alghalibi 2000, Xie *et al.* 2001, and Keay *et al*. 2006.

In a study in Bangladesh, antecedent ocular trauma was reported by 35% of patients with mycotic keratitis and 52% of patients with bacterial keratitis (Dunlop *et al*. 1994). Data derived from the retrospective case-control study in Singapore (Wong *et al*. 1997), suggested that mycotic keratitis (principally due to *Aspergillus* spp. and *Fusarium* spp.) was more likely to be related to mechanical ocular trauma and bacterial keratitis (principally due to *Pseudomonas aeruginosa*) was more likely to be related to contact lens wear and preexisting ocular diseases. Preexisting inflammatory ocular diseases were less frequently seen in mycotic keratitis than in bacterial keratitis, but systemic immunosuppressive conditions appeared to be of equal significance in both mycotic and bacterial keratitis. However, in the current study, both diabetic and immunosuppression positive patients had keratitis due to fungal agents, while the hypertensive patients and contact lens wearers had keratitis due to bacterial agents.

Table 1: Collective data of keratitis cases due to bacterial and/or fungal species

Types of keratitis and organisms isolated	Number of isolates	Percentage of isolates (%)
Bacterial keratitis (50 cases)		
Staphylococcus aureus Rosenbach	30	60
Staphylococcus sp.	15	30
Pseudomonas aeruginosa (Schroeter) Migula	2	4
Klebsiella pneumoniae (Schroeter) Trevisan	1	2
Actinomycete sp.	1	2
Micrococcus luteus (Schroeter) Cohn	1	2
Escherichia coli (Migula) Castellani and Chalmers	1	2
Fungal keratitis (30 cases)		
Aspergillus flavus Link	10	33.3
Aspergillus terreus Thom	4	13.3
Aspergillus niger van Tieghem	3	10.0
Candida albicans (Robin) Berkhout	2	6.7
Cladosporium cladosporioides (Fresenius) de Vries	2	6.7
Aspergillus fumigatus Fresenius	1	3.3
Candida kruisii (Kochova-Kratochvilova & Ondrusova) A. Meyer & Yarrow	1	3.3
Fusarium oxysporum Schlechtendal	1	3.3
Fusarium proliferatum (Matsushima) Nirenberg	1	3.3
Fusarium solani (Martius) Saccardo	1	3.3
Penicillium chrysogenum Thom	1	3.3
Sterile mycelia	1	3.3
Cochliobolus spicifer Nelson	1	3.3
Trichderma hamatum (Bonorden) Bainier	1	3.3
Mixed keratitis (5 cases)		
Staphylococcus aureus Rosenbach	4	80
Klebsiella pneumoniae (Schroeter) Trevisan	1	20
Staphylococcus sp.	1	20
Aspergillus flavus Link	2	40
Cladosporium cladosporioides (Fresenius) de Vries	1	20
Fusarium verticillioides Sheldon	1	20
Stemphylium botryosum Wallroth	1	20

*This figure is due to the mixed cases from which a fungal and a bacterial species were found.

Table 2: Age and gender (M: male; F: female) distribution of 115 cases of keratitis in Upper Egypt

Age Group (years)	Total keratitis				Bacterial keratitis				Fungal keratitis				Mixed keratitis				Negative			
	M	F	Total	%	M	F	Total	%	M	F	Total	%	M	F	Total	%	M	F	Total	%
< 1 year	-	1	1	0.9	-	1	1	2	-	-	-	-	-	-	-	-	-	-	-	-
1-10	4	2	6	5.2	2	-	2	4	-	1	1	3.3	1	1	2	40	1	-	-	-
11-20	4	2	6	5.2	1	-	1	2	2	1	3	10	-	1	1	20	1	-	1	3.3
21-30	7	2	9	7.8	4	-	4	8	2	1	3	10	-	-	-	-	1	1	2	6.7
31-40	13	4	17	14.8	6	2	8	16	4	-	4	13.3	-	1	1	20	4	1	5	16.7
41-50	15	8	23	20	7	6	13	26	1	1	2	6.7	-	-	-	-	7	1	8	26.7
51-60	16	8	24	20.9	3	1	4	8	7	5	12	40	1	-	1	20	5	1	7	23.3
61-70	15	4	19	16.5	8	2	10	20	3	1	4	13.3	-	-	-	-	4	1	5	16.7
71-80	7	2	9	7.8	4	2	6	12	1	-	1	3.3	-	-	-	-	2	-	2	6.7
81-90	1	-	1	0.9	1	-	1	2	-	-	-	-	-	-	-	-	-	-	-	-
Total	82	33	115		36	14	50		20	10	30		2	3	5		25	5	30	
% Total	71.3	28.7			72	28			66.7	33.3			40	60			83.3	20		

Table 3: Incidence of keratitis in relation to occupation of patients

Occupation	Total		Bacterial		Fungal		Mixed		Negative	
	No.	%	No.	%	No.	%	No.	%	No.	%
Farmers	46	40	21	42	12	40	1	20	12	40
Housewives	19	16.5	8	16	7	23.3	1	20	3	10
Unknown	17	14.8	8	16	3	10	-	-	6	20
Workers	9	7.8	4	8	4	13.3	-	-	1	3.3
Students	8	7.0	2	4	2	6.7	2	40	2	6.7
Retired	7	6.1	4	8	1	3.3	-	-	2	6.7
Children	4	3.5	2	4	1	3.3	1	20	-	-
Drivers	2	1.7	-	-	-	-	-	-	2	6.7
Employees	2	1.7	-	-	-	-	-	-	2	6.7
Soldiers	1	0.9	1	2	-	-	-	-	-	-
Total/Percentage per category	115	100	50	43	30	26	5	4	30	26

Table 4: Incidence of keratitis in relation to risk factors

Risk factor	Total		Bacterial		Fungal		Mixed		Negative	
	No.	%	No.	%	No.	%	No.	%	No.	%
1- Trauma:	73	63.4	31	62	23	73.3	4	80	15	50
a- Plant	57	49.6	26	52	18	60	3	60	10	33.3
b- Foreign bodies	8	7	2	4	3	10	1	20	2	6.7
c- Unknown	8	7	3	6	2	6.7	-	-	3	10
2- Unknown factors	22	19.1	14	28	1	3.3	-	-	7	23.3
3- ACS	8	7	3	6	2	6.7	1	20	2	6.7
4- Diabetic patient	5	4.3	-	-	3	10	-	-	2	6.7
5- Hypertension	3	2.6	1	2	-	-	-	-	2	6.7
6- Contact lens wear	2	1.7	1	2	-	-	-	-	1	3.3
7-ISP	2	1.7	-	-	1	3.3	-	-	1	3.3
Total/Percentage per category	115	100	50	43	30	26	5	4	30	26

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