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## Efficient role of VITEK systems in rapid identification of different corneal ulcer infectious microorganisms during an updated survey in Egypt

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### Abstract

For one year survey to study predisposing factors of microbial keratitis, a total of 632 patients were attending Ophthalmology Department, Tanta University Hospital, where 7 patients had infection in both eyes so total infected eyes were 639. There were 10 cases with mixed infection, so the total isolates obtained from 639 infected eyes were 649 isolates (362 bacterial isolates = 55.8% out of total isolates and 287 fungal or yeast isolates = 44.2% out of total isolates). The most common predisposing factors for infectious keratitis were ocular trauma. Although vegetative trauma was the most common risk factor for microbial keratitis, contact lens wearing, use of steroids, dry eyes, were important risk factors for keratitis. VITEK® systems were used for more accurate identification of microbial keratitis causative agents and decreasing the turnaround time. The Mean time of identification turnaround time was for filamentous fungi (25 minutes), bacteria (4:51 hours) and yeast (17:59 hours). It was observed that the predominant bacterial isolate was *Staphylococcus aureus* (131 isolates 20% of total isolates) followed by *Pseudomonas aeruginosa* (81 isolates 12% of total isolates). The predominant yeast isolate was *Candida albicans* (58 isolates/ 9% of total isolates). The predominant filamentous fungal isolate was *Aspergillus flavus* (44 isolates/ 7% of total isolates).

**Keywords:** Keratitis, VITEK®, MALDI-TOF, predisposing factors, demographic data

### 1. Introduction

The ocular ailment known as keratitis is characterized by corneal inflammation. The transparent layer covering the cornea acts as a physical barrier and is in charge of causing light to be refracted onto the retina. According to Cleveland (2018) [4], it is made up of five layers, the outermost of which is the corneal epithelium, which works with the tear film to clean and shield the eye. The epithelium is made up of five to seven layers of non-keratinized cells and has a thickness of roughly fifty micrometers (Voss *et al.*, 2021) [25]. Ocular morbidity may result from inflammatory cells invading the corneal epithelium, particularly in cases when the infiltration is deeper.

Trauma to the eyes can damage the corneal epithelium and impair its defenses, which can lead to a localized opportunistic infection. When a local infection starts at the epithelium, leukocytes and macrophages are drawn in. Together with the pathogen, these inflammatory cells release hydrolases and proteases that have harmful effects on vision function (Singh *et al.*, 2021) [18].

Even though microbial keratitis is a major contributor to corneal blindness and visual impairment, particularly in developing nations (Whitcher *et al.*, 2001) [26], the current research region lacks prior epidemiological reports for various forms of microbial keratitis. In this study, the epidemiological profile and key risk factors for microbial keratitis at Tanta University Ophthalmology Hospital in Egypt were to be described.

In addition to microbiological assessment, the clinical foundation is used to diagnose microbial keratitis (Ung *et al.*, 2019) [23]. Global variations exist in the microbiological profile of microbial keratitis. VITEK® systems offer quick and accurate characterization of various microorganisms, which is a critical step in the appropriate prevention and control of diseases in medical diagnostics and is currently a major source of worry (El-Behiry *et al.*, 2014) [5].

## 2. Materials and Methods

### 2.1 Collection of samples and culture techniques

**Clinical diagnosis of corneal ulcers:** A study lasting for one year was conducted in 2023, from January to December. Samples were obtained during routine follow-up visits for patients admitted to Tanta University's Inpatient Department of Ophthalmology Hospital, Egypt; and twice a week visits to the Outpatient Clinic. Photographs were taken of the patient's symptoms. Samples were taken from people with various types of corneal ulcers who had been clinically identified with them.

#### 2.1.1 Selection of cases

Selected patients were subjected to the following parameters for more accurate data and sample collection in the present study, that were previously collected by ophthalmology consultants (El-Esawy *et al.*, 2024) [6].

##### 2.1.1.1 Inclusion criteria

- Patients with infectious central corneal ulceration presenting to Ophthalmology Outpatient Clinic and Inpatient Department of Ophthalmology Hospital, Tanta University.
- Patients will be included after examination and clinical diagnosis of corneal ulcer.

##### 2.1.1.2 Exclusion criteria

- Typical viral and acanthamoebal ulcers were excluded.
- Moorens ulcer.
- Sterile neutrotrophic ulcers.
- Interstitial ulcers.

##### 2.1.1.3 Ethical consideration

- The ethical rules issued by Tanta University's ethical committee (approval code: IACUC-SCI-TU-0101) and any subsequent changes or comparable ethical guidelines were adhered to by all procedures involving human subjects carried out in this study. Each individual taking part in the research provided informed consent.
- There was privacy for participants and confidentiality of the data.
- Any unanticipated risks that arose throughout the study were promptly disclosed to participants and the ethical committee.
- After informing the patients about the procedure, an informed written consent was taken.

#### 2.1.2 History taking

Age, gender, family crowding, occupation, ocular treatment, house type, sewage disposal, traumatic agents and clinical profile were recorded.

#### 2.1.3 Samples investigations

After administering an anesthetic topical eye drop (Boxinate, Alex. Pharma, Egypt), corneal scrapings were collected from the ulcer's edge using a sterile No. 15 blade, and their non-antimicrobial activity was examined. Two glass slides were scraped, one for KOH mounting and the other for Gram staining (Margo, and Brinser, 1987) [13].

### 2.2 Identification of bacterial and yeast isolates

The protocol of sample preparation and loading on different VITEK systems in the present study was adopted according to the methodology of the system manual for both bacterial and yeast types (Versalovic *et al.*, 2011) [24], as follows.

#### 2.2.1 Culture Requirements

A fresh inoculum was prepared from 12 to 48 hours age culture, with inoculum density of 0.50 to 0.63 McFarland Standard, incubated at 35 °C to 37 °C. Age of suspension before loading on the instrument shouldn't exceed 30 minutes (Iversen *et al.*, 2008) [11].

#### 2.2.2 Materials

The VITEK® 2 apparatus cards were comprehensive methods for routine identification testing of most clinically relevant Gram positive, Gram negative, and yeast organisms. The following supplies were needed: VITEK® 2 Card, VITEK® 2 DensiCHEK™ Plus Kit, DensiCHEK™ Plus Standards Kit, VITEK® 2 Cassette, Sterile saline (aqueous 0.45% to 0.50% NaCl, pH 5.5 to 7.0), 12 mm x 75 mm disposable test tubes made of clear plastic (Polystyrene), both the nutrition medium and sterile sticks or swabs (Iversen *et al.*, 2008) [11].

#### 2.2.3 Procedure

A test tube made of clear plastic (polystyrene) measuring 12 mm by 75 mm was aseptically filled with three milliliters of sterile saline (aqueous 0.45% to 0.50% NaCl, pH 5.5 to 7.0). The saline tube was created and sufficient morphologically comparable colonies were put into it. Using a calibrated VITEK® 2 DensiCHEK™ Plus or VITEK® 2 DensiCHEK™, a homogenous bacterial solution was made with a density comparable to a McFarland No. 0.50 to 0.63 (or, for yeast, McFarland No. 1.80 to 2.20). The age of suspension cannot be more than 30 minutes prior to the vaccination card. After inserting the card and suspension tube into the cassette, the tape was loaded into the instrument (Versalovic *et al.*, 2011) [24].

#### 2.2.4 Methods used for filamentous fungi identification

Time of Flight for Matrix-Assisted Laser Desorption Ionization (MALDI TOF) Mass Spectrometry, the automatic identification of filamentous fungus was done using the VITEK®MS System. The following was how the cultivation process was carried out: After the fungal colony had grown sufficiently on a Sabouraud's dextrose agar plate, its periphery was swabbed and suspended in API suspension media. Add 900 µL of pure ethanol and vortex. After centrifuging the tube for two minutes at 10000 rpm, the supernatant was disposed of.

After adding 40 µL of 70% formic acid to the pellet, it was vortexed. Once added, 40 µL of 100% acetonitrile was vortexed. The tube was centrifuged at 10000 rpm for two minutes. After adding 1 µL of the supernatant to the target slide, it was allowed to dry at room temperature. After applying 1 µL of the CHCA matrix to the sample site, it was allowed to dry at room temperature (Suh *et al.*, 2013) [20].

## 3. Results

The corneal ulcers are more common among patients aged between 40- 50 years old (161 cases) which represent 25.47% of total cases (632 cases), and the lower cases number recorded in the patients aged less than ten years old, as revealed in Table (1). Females (342 cases = 54.11% out of total corneal ulcers) were more affected than male (290 cases = 45.89% out of total corneal ulcers) as shown in Table (2). The prevalence of corneal ulcers in family crowding  $\geq 4$  (372 cases = 58.86% out of all cases) was more than Family crowding  $< 4$  (260 cases represent 41.14% out of all cases) in the study, as in Table (3).

From Table (4), it was observed that the majority of cases (430 cases = 68.04% out of total cases) did not receive any

ocular treatment in previous year before the infection. (71 cases = 11.23% out of total cases) received steroids and (42 cases = 6.65% out of total cases) received antibacterial antibiotics. Antifungal drugs administered by 35 cases (5.54% out of total cases). While 54 cases (8.54% out of total cases) treated with lubricant eye drops due to suffering from dry eye problems.

From Table (5), it was observed that the patients who live in rural houses (330 patients = 52.22% out of total patients) are more susceptible to microbial keratitis than who live in urban houses (302 patients = 47.78% out of total patients). From Table (6), it was observed that the patients who used closed pipe network (388 patients = 61.39% out of total patients) were more affected by microbial eye infections than who use conservancy (244 patients = 38.61% out of total patients).

From Table (7), it was observed that the highest rate of infections was recorded in housewives (205 patients = 32.4% out of total patients), followed by farmers (170 patients = 26.9% out of total patients), then handy professionals and indoor employees that represents 119 and 113 patients with the percentage of 18.8% and 17.9%. The lowest corneal ulcers incidence was presented in students (25 patients = 4.0% out of total patients).

Out of total patients, 491 cases had traumatic agents, representing 77.69%. Table (8) revealed that the major cause of microbial keratitis is trauma, the traumatic agents varies among the cases. Vegetative matters (168 cases = 26.58% out of total traumatic agent), Sand/dust (62 cases = 9.81% out of total traumatic agent), Contact lense (132 cases = 20.89% out of total traumatic agent), Ocular surgery (48 cases = 7.59% out of total traumatic agent), Metallic trauma (43 cases = 6.80% out of total traumatic agent), Animal scratches (22 cases = 3.48% out of total traumatic agent), Insects (11 cases = 1.74% out of total traumatic agent) and Fingernails (5 cases = 0.79% out of total traumatic agent).

Within the total 632 patients, 7 patients had infection in both eyes so total eyes infected are 639. From Table (9), the clinical profile of the corneal ulcers revealed that the only right eye infections were 369 (57.7%), more than the only left eye infection 263 (41.2%) and the number of patients who had infections in both eyes are seven patients (1.1%). The site of corneal lesions in the center of cornea (365 cases = 57.1% out of total corneal ulcers) more than of peripheral infection (274 cases = 42.9% out of total corneal ulcers). The size of microbial corneal ulcers which  $\geq 3$  mm were (380 infections = 59.5% out of total corneal ulcers), more than infection size  $< 3$  mm (259 infections = 40.5% out of total corneal ulcers). The medical condition of hypopyon formation was considered as a distinguishable sign for resistant ulcers due to the formation of this precipitate during the prolonged immune response of host tissues against the formed ulcers; this was observably represented in the present survey for 294 infected eyes (= 46% out of total corneal ulcers) with different types of the studied microbial infections. Time interval between the first pain and the appearance of ulcer lesion on the patient's eye was major evidence for the severity, duration of ulcer and microbial offensive force, that affect the degree of damage of corneal tissues, microbial growth rate and the chance of corneal healing on the appropriate treatment. There were 246 infections (38.5%), reported the observable superficial ulcer signs within 1 week interval after the first pain of patient; 221 infections (34.6%) were reported ulcer signs within the second week after the first pain; other 125 infections (19.6%) reported more ulcer complications within

the third week after the first pain; while 47 infections (7.3%) showed more deep corneal damage and vigorous complications within the fourth week of different microbial infection types, as revealed in Table (9).

Total 632 patients, 7 patients had infection in both eyes, 10 cases had mixed infection, so the total isolates obtained from 639 infected eyes are 649 isolates (362 bacterial isolates = 56% out of total isolates and 287 fungal and yeast isolates = 44% out of total isolates), 41% of ulcers presented in the summer, 23% in the fall, 7% in the winter, and 29% in the spring, as presented in Table (10).

Some representative symptoms of the different types of microbial ulcers on the infected eyes were summarized in Photo (1), showing the formation of hypopyon, scars, opacity and other complications of the infected corneal tissues, severely impairing the vision of the patient.

From Table (11), it was observed that the predominant bacterial isolate was *Staphylococcus aureus* (131 isolates, 20% of total isolates) followed by *Pseudomonas aeruginosa* (81 isolates, 12% of total isolates). The predominant yeast isolate was *Candida albicans* (58 isolates, 9% of total isolates). The predominant filamentous fungi pathogen isolated was *Aspergillus flavus* (44 isolates, 7% of total isolates). Among all yeast isolates identified, *Candida albicans* was represented by 61% of total identified yeasts; the *Staphylococcus aureus* was represented by 36.2% of total bacterial isolates; and *Aspergillus flavus* recorded percentage of 23% of total fungal isolates.

Identification turnaround time varied significantly in the three groups, in bacteria group, the mean time for identification using VITEK® 2 compact is 4.51 h, whereas in yeast group the mean time is 17.59 h. Identification turnaround time for filamentous fungi using VITEK® MS MAIDI TOF was dramatically shorter than bacterial and yeast identification due to direct hyphal breakage step as an accelerating technique for biochemical testing. The mean time for fungal identification including sample preparations is 25 minutes as shown in Table (12).

#### 4. Discussion

The present results showed that the corneal ulcers were more common among patients aged between 40- 50 years old (161 cases) which represent 25.47% of total cases (632 cases), that was supported by Politis *et al.* (2016) [15], who reported that the mean age of patients with corneal ulcers is (47.0 ± 25.2) and the bacterial corneal infections were more than fungal infections. A study conducted at Ain Shams University (Anwar *et al.*, 2022) [1] showed that the patients' ages ranged from 29 to 71 years old, with a mean of 46.67 ± 10.90 years. In the current survey, the age group with the highest frequency of age presentations (36.7%) was 41 to 50 years old, that also agreed with Sakr *et al.* (2023) [17], who conducted a study from 2017 to 2022 at Alexandria University and found that the mean age was 40.4 years and ocular trauma was the most common predisposing factor for microbial keratitis.

In the present study, females (342 cases = 54.11% out of total corneal ulcers) were more affected than males (290 cases = 45.89% out of total corneal ulcers). Interestingly, one study from Nepal reported a higher proportion of females compared to males (Suwal *et al.*, 2016) [22], whilst other studies from Nepal reported male preponderance (Sitoula *et al.*, 2015) [19]. It was undefined reason why there is a difference; it could be because of various socioeconomic characteristics, health-seeking habits or changing in lifestyles.

In the present survey, Traumatic agents represented 77.69%



out of total patients in the study. The major cause of microbial keratitis is trauma, the traumatic agents varied among the cases. Vegetative matters (168 cases = 26.58% out of total traumatic agent) followed by Contact lens usage (132 cases = 20.89% out of total traumatic agent) and Ocular surgery (48 cases = 7.59% out of total traumatic agent). It is observed that the majority of cases (430 cases = 68.04% out of total cases) did not receive any ocular treatment in previous year before the infection. (71 cases = 11.23% out of total cases) receive steroids followed by (54 cases = 8.54% out of total cases) treated with lubricant eye drops due to suffering of dry eyes problems. A study conducted at Ain Shams University (Anwar *et al.*, 2022) [1] revealed that trauma was the most common predisposing factor (46.6%), followed by steroids (20%) and post-operative (6.7%).

In Karimizadeh *et al.* (2022) [12] prolonged survey, 35 appropriate studies from six nations (Iraq, Egypt, Turkey, Iraq, Saudi Arabia and Oman) were included in a meta-analysis study. Combining the study results allowed for the estimation of the pooled prevalence of keratitis. Trauma was identified as the main risk factor in studies from Saudi Arabia and Egypt, according to the random-effects meta-analysis (I, 2 = 98.88%,  $p < 0.001$ ). Egypt had the highest rate of trauma prevalence (51%) while Iran had the lowest rate (0.3%). In Turkey, 50% of contact lens wearers utilize them improperly; 30% have had eye surgery in the past; and 22.7% of Egyptians use topical steroids.

The size of microbial corneal ulcers which  $\geq 3$  mm were (380 infections = 59.5% out of total corneal ulcers) was more incident than infection size  $< 3$  mm (259 infections = 40.5% out of total corneal ulcers). Hypopyon formation in 294 infections (= 46% out of total corneal ulcers) in the present study was significantly related to the more resistant fungal, bacterial and mixed ulcers. This agreed with the findings of the study published by Chidambaram *et al.* (2018) [2], who reported that *Aspergillus* species and bacterial keratitis were more associated with hypopyon. Another study conducted at Assiut University revealed that hypopyon represent (31.3%) of total cases, which were more related with prolonged resistant ulcers (Hamza *et al.*, 2022) [9].

The severity of microbial ulcer, degree of tissue damage of the infected cornea and the strategy of treatment protocol were basically dependent on the time interval between first patient pain sensation and the appearance of the first visible sign of ulcer to start the appropriate diagnosis and treatment. That ensures the importance of rapid and early detection of microbial ulcers and identification of their causative agents as a main aim for the present study. In the present study, most cases were diagnosed with the ulcer signs within the first week after pain recording (246 cases = 38.5%), followed by 221 infections (34.6%) within the second week, then 125 infections (19.6%) within the third week, while only 47 infections (7.3%) reported more complications within the fourth week keratitis sensation. Another study supported the present survey findings, as Wuletaw *et al.* (2021) [16] recorded observable ulcer signs for 43.3% of corneal ulcer patients within 1-7 days of first patient pain, while the lower number of patients presented their ulcer appearance after more than 30 days of first pain sensation (only 13.3%).

In the present survey, 41% of ulcers presented in the summer, 23% in the Autumn, 7% in the winter, and 29% in the spring. These results agreed with Gorski *et al.* (2016) [7], who recorded that infectious keratitis occurred more frequently in the summer. Warmer temperatures, more

humidity and increased ocular exposure to water were potential causes of this heightened summer appearance.

The present results revealed that using VITEK® 2 compact system decreased the mean turnaround time for identification of bacterial isolates to 4.51 hours, which was in agreement with Hernández-Durán (2017) [10], who conducted a comparison between automatic bacterial identification devices (VITEK® 2 compact and MicroScan system) and found that with the VITEK® 2 / MicroScan systems, the median time for conclusive identification of Gram-negative Enterobacteriaceae was 5.1-12.33 hours, for non-Enterobacteriaceae, it was 6.8-26.7 hours and for Gram-positive cocci, *Staphylococcus spp.*, it was 5.6-19.92 hours and Enterococcus spp., 9.6-16.8 hours.

In the present study, the mean turnaround time for identification of yeast isolates by VITEK 2™ was 17.59 hours. This is in accordance with studies carried out by Melhem *et al.* (2014) [14] and Graf *et al.* (2000) [8], where the TAT by VITEK 2™ was 18 hr. and 15 hr. In a study by Sukanya *et al.* (2021) [21], the average turnaround time was discovered to be 15.42 hours, which was once more less than what the traditional method requires. The decreased TAT by VITEK 2™ was statistically significant ( $p < 0.01$ ) when compared, which was in line with other investigations. In the present study, Identification turnaround time for filamentous fungi using VITEK® MS MAIDI TOF is dramatically shorter than bacterial and yeast identification. The mean time for fungal identification including sample preparations is 25 minutes. These results agreed with Chun *et al.* (2019) [3], who reported that, with confidence ratings as high as 99.9%, MALDI-TOF MS successfully identified pathogens from intraocular samples through direct analysis. The processing of the material took twenty minutes. Applying a single spot to the target plate took thirty seconds. Using MALDI-TOF MS, a single spot on the target plate could be analyzed in less than 60 seconds. After analysis, the data were accessible in less than five minutes. From sample preparation to result acquisition, the entire process of pathogen identification using MALDI-TOF MS took less than 30 minutes for a single sample.

**Table 1:** Distribution of infectious corneal ulcers among different age groups

Age group (years)	Total cases	% of incidence
< 10	2	0.32
$\geq 10-20<$	63	9.97
$\geq 20-30<$	81	12.82
$\geq 30-40<$	107	16.93
$\geq 40-50<$	161	25.47
$\geq 50-60<$	125	19.78
$\geq 60-70<$	83	13.13
$\geq 70$	10	1.58
Total patients	632	100%

**Table 2:** Distribution of infectious corneal ulcers among different gender groups

Gender type	No. of cases	% of incidence
Male	290	45.89%
Female	342	54.11%
Total patients	632	100%

**Table 3:** Effect of family crowding index on the incidence of infectious corneal ulcers

Family crowding	No. of cases	% of incidence
$\geq 4$	372	58.86%
$< 4$	260	41.14%
Total patients	632	100%

**Table 4:** Effect of previous ocular treatment on the distribution of corneal infections

Type of treatment	No. of cases	% of incidence
No previous treatment	430	68.04%
Steroids	71	11.23%
Antibacterial	42	6.65%
Antifungal	35	5.54%
Lubricant eye drops	54	8.54%
Total patients	632	100%

**Table 5:** Distribution of infectious corneal ulcers among different house types

House type	No. of cases	% of incidence
Rural	330	52.22%
Urban	302	47.78%
Total patients	632	100%

**Table 6:** Effect of sewage disposal type on the incidence of infectious corneal ulcers

Sewage disposal	No. of cases	% of incidence
Closed pipe network	388	61.39%
Conservancy	244	38.61%
Total patients	632	100%

**Table 7:** Distribution of microbial corneal ulcers among different occupational careers of the studied patients

Occupation career	No. of cases	% of incidence
Housewives	205	32.4%
Farmers	170	26.9%
Handy professionals	119	18.8%
Indoor employees	113	17.9%
Students	25	4.0%
Total patients	632	100%

**Table 8:** Types of traumatic agents recorded in the present survey

Traumatic agents	No. of cases	% of incidence
Vegetative matters	168	26.58%
Fingernails	5	0.79%
Insects	11	1.74%
Animal scratches	22	3.48%
Metallic trauma	43	6.80%
Contact lens usage	132	20.89%
Ocular surgery	48	7.59%
Sand/dust	62	9.81%
Total traumatic agents	491	77.69%
Non traumatic agents	141	22.31%
Total patients	632	100%

**Table 9:** Clinical profile of infected corneas in the present survey

Parameters	Categories	No. of cases	% of incidence
Infected eye	Left eye only	263	57.7%
	Right eye only	369	41.2%
	Both eyes	7	1.1%
Site of corneal lesions	Central	365	57.1%
	Peripheral	274	42.9%
Size of ulcer	< 3 mm	259	40.5%
	≥ 3 mm	380	59.5%
Hypopyon formation	Present	294	46.0%
	Absent	345	54.0%
Time interval between first pain and first ulcer sign appearance	1-7 days	246	38.5%
	8-14 days	221	34.6%
	15-21 days	125	19.6%
	21-28 days	47	7.3%
Total infected eyes for each parameter		639	100%

**Table 10:** Incidence of infectious corneal ulcers during the four seasons of the present survey

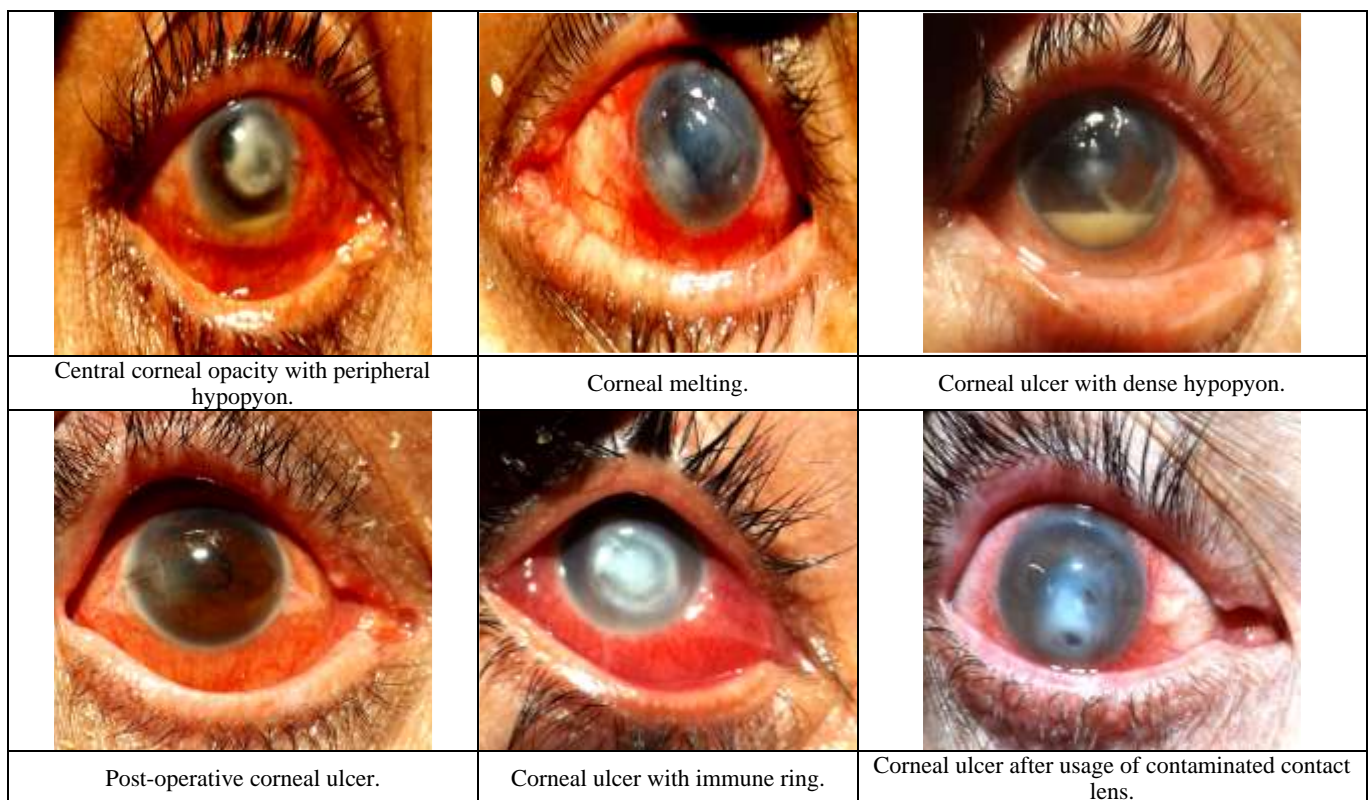
Type of microbial infection	Winter	Spring	Summer	Autumn	Total
Bacterial only	36	104	153	59	352
Fungal or yeast only	9	79	101	88	277
Mixed infections	0	2	5	3	10
Total bacterial isolates	36	106	158	62	362
Total Fungal and yeast isolates	9	81	106	91	287
Total isolates	45	187	264	153	649

**Table 11:** Incidence of different microorganisms, isolated from microbial keratitis and identified by VITEK systems during the present study

Bacteria		Yeast		Filamentous fungi	
Gram positive bacteria		<i>Candida albicans</i>	58	<i>Aspergillus niger</i>	39
<i>Staphylococcus aureus</i>	131	<i>Candida glabrata</i>	7	<i>Aspergillus flavus</i>	44
<i>Staphylococcus haemolyticus</i>	21	<i>Candida rugosa</i>	3	<i>Aspergillus fumigatus</i>	26
<i>Staphylococcus epidermidis</i>	15	<i>Candida utilis</i>	3	<i>Fusarium solani</i>	13
<i>Streptococcus pneumoniae</i>	12	<i>Candida famata</i>	4	<i>Fusarium oxysporum</i>	5
<i>Enterococcus faecalis</i>	24	<i>Candida haemulonii</i>	2	<i>Penicillium lanosum</i>	14
<i>Enterococcus durans</i>	2	<i>Candida lipolytica</i>	1	<i>Penicillium spinulosum</i>	10
<i>Staphylococcus hominis</i>	6	<i>Candida krusei</i>	3	<i>Penicillium citrinum</i>	3
<i>Streptococcus canis</i>	1	<i>Candida tropicalis</i>	7	<i>Acremonium potronii</i>	1
<i>Streptococcus viridans</i>	2	<i>Cryptococcus laurentii</i>	3	<i>Bipolaris spicifera</i>	2
<i>Corynebacterium striatum</i>	1	<i>Saccharomyces cerevisiae</i>	1	<i>Fusarium dimerum</i>	4
<i>Streptococcus agalactiae</i>	1	<i>Saprochaete capitata</i>	2	<i>Trichophyton schenleinii</i>	1
<i>Streptococcus pyogenes</i>	1	Total yeast isolates	94	<i>Mucor fuscus</i>	2
<i>Staphylococcus saprophyticus</i>	2			<i>Trichophyton verucosum</i>	1
Gram negative bacteria				<i>Mucor fuscus</i>	2
<i>Pseudomonas aeruginosa</i>	81			<i>Cladosporium Sphearospermum</i>	2
<i>Serratia marcescens</i>	1			<i>Curvularia lunata</i>	2
<i>klebsiella pneumoniae</i>	34			<i>Alternaria alternata</i>	14
<i>Enterobacter bacillus</i>	10			<i>Microsporium canis</i>	7
<i>Proteus penneri</i>	4			Total fungal isolates	193
<i>Proteus mirabilis</i>	2				
<i>Escherichia coli</i>	8				
<i>Acitinobacter haemolyticus</i>	1				
<i>Moraxella osloensis</i>	1				
<i>Nocardia asteroides</i>	1				
Total bacterial isolates	362				

**Table 12:** Turnaround time for identifications of different microbial types, using VITEK® systems, compared to traditional methods of identification

Organism	Traditional culture methods turnaround time (mean)	VITEK® systems identification turnaround time (mean)
Bacteria	24 h	4.51 h
Yeast	48 h	17.59 h
Filamentous fungi	7 days	25 minutes



**Photo 1:** Representative cases of different symptoms of microbial corneal ulcers



## 5. Conclusion

Trauma was the most predisposing factor for microbial corneal ulcers in this study. Using VITEK® 2 compact and VITEK® MS MALDI-TOF with less turnaround time for detection of microorganism may help in early detection and proper prognoses of infected corneal ulcer cases.

**6. Conflict of interest:** There is no conflict of interest.

**7. Financial interest:** There was no fund for this research article.

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