

## ORIGINAL ARTICLE

# *Pseudomonas* Keratitis Predominance in Government Hospitals in Johor, Malaysia

Vidhubala Balasegar<sup>1,3</sup>, Rafidah Md Saleh<sup>2</sup>, Leslie Thian Lung Than<sup>1</sup>, Azmiza Syawani Jasni<sup>1</sup>, Rosni Ibrahim<sup>1</sup><sup>1</sup> Department of Medical Microbiology, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia<sup>2</sup> Department of Ophthalmology, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia<sup>3</sup> Department of Pathology, Sultan Ismail Hospital, Jalan Mutiara Emas Utama, Taman Mount Austin, 81100 Johor Bahru, Johor, Malaysia**ABSTRACT**

**Introduction:** Keratitis is an eye-threatening condition, which requires immediate treatment, due to the rapid progression when the infection caused by *Pseudomonas aeruginosa*. This study aimed to describe the socio-demographic factors and risk factors related to *Pseudomonas* keratitis in selected six government hospitals in Johor, Malaysia. **Methods:** An 18 months cross-sectional study was conducted among bacterial keratitis patients from six government hospitals offering ophthalmology services in Johor, Malaysia. The cases were confirmed through a positive culture of *Pseudomonas aeruginosa*. All data on socio-demographic factors, risk factors, and symptoms were recorded using a data collection form and analysed using the Statistical Package for Social Sciences (SPSS) software version 26. **Results:** Fifty-five patients were diagnosed with *Pseudomonas* keratitis in this study. The majority of patients were males (65.5%) with a mean age of 41.22 years old. The majority of patients were of Malay ethnicity (60.0%). More than a quarter of cases are involved in office-oriented jobs (n=14, 25.5%). The major risk factors in this study were contact-lens usage (n=25, 45.5%), trauma (n=20, 36.4%), ophthalmic steroid usage (n=16, 29.1%), ocular surface disease (n=12, 21.8%) and previous eye surgery (n=4, 7.3%). This study found underlying medical illness (p=0.036) and ocular surface disease (p=0.051) are significantly associated with *Pseudomonas* keratitis. **Conclusion:** The present study is the first study in Malaysia focusing on *Pseudomonas* keratitis. This study provides additional information on the epidemiology data of keratitis in Malaysia. Future studies shall be extended to all government hospitals in Malaysia to obtain better insight on the disease burden of keratitis especially on *Pseudomonas* keratitis.

Malaysian Journal of Medicine and Health Sciences (2024) 20(2): 4-10. doi:10.47836/mjmhs.20.2.2

**Keywords:** Contact-lens; Keratitis; *P.aeruginosa*; Risk factors; Socio-demographic**Corresponding Author:**

Rosni Ibrahim, MPath

Email: rosni.ibrahim@upm.edu.my

Tel: +603-97692363

or even severe cases leaving scar even after the ulcer healed. Due to the severity of the disease, *Pseudomonas* keratitis gained more attention among ophthalmologists compare to keratitis caused by other organisms [4].

**INTRODUCTION**

*Pseudomonas* keratitis is a severe pathological manifestation that can lead to blindness, although better treatment options are available today [1,2]. In particular, keratitis caused by *Pseudomonas aeruginosa* cause severe disease presentation than other bacteria due to the fact that *Pseudomonas aeruginosa* produces various endotoxin and enzymes which can damage the cornea directly or leave bacterial products which aggravates the host immune response even after the bacteria no longer multiplying in cornea [3]. Although *Pseudomonas* keratitis easily identified yet the ulcer is far more difficult to treat and often result in poor visual outcome

*Pseudomonas aeruginosa* has a unique ability to adhere to living cells and also non-living materials such as contact-lenses [5]. The bacterium also produces various virulence factors such as ExoU, ExoS, alkaline protease and possess flagella that enabling it to move reside in the cornea without being destroyed by the natural corneal immune system. Virulence factors such as ExoU and ExoS was described in previous study as important factors in *Pseudomonas* keratitis [6,7].

The prevalence of *Pseudomonas* keratitis in Malaysia presented since the first study in 2005 [8] till now [9]. Moreover, the pattern of socio-demographic factors and risk factors are similar across studies in Malaysia in

which gender males aged between 40-60 years old and ethnic Malay are the most affected group. In addition, similar risk factors were observed, like contact-lens usage to be the major risk factor in microbial keratitis associated with *Pseudomonas aeruginosa*. Globally, some literature reported that the most common isolated organism for bacterial keratitis was *Pseudomonas aeruginosa* [10–15].

The aim of this study was to determine the proportion of *Pseudomonas* keratitis, to determine the socio-demographic, the risk factors, the symptoms and the association of these parameters with *Pseudomonas* keratitis in patients attending selected government hospitals in Johor, Malaysia. Moreover, the data from this study aimed to provide information to relevant policy makers on strengthening the measures on the prevention of eye injuries and to create awareness on the importance of eye protection among target groups in the community.

## MATERIALS AND METHODS

### Research Design

A cross-sectional study was carried out in selected government hospitals in Johor, Malaysia. The study duration was from June 2019 to December 2020. All patient information was collected using a data collection forms that collected data on patients' gender, age, ethnicity, occupation, risk factors and also symptoms.

### Study Location

Johor, Malaysia's southernmost state, occupies a land area of 19,166 km<sup>2</sup>. The population of Johor was estimated at 3.76 million as of 2019 [16]. The state has ten districts. Johor Bahru, the capital city of Johor, is home to nearly 42% (1.6 million) of the state population. The state has twelve government hospitals, with six hospitals offering ophthalmology services. The hospitals which was selected for this study was Hospital Sultanah Aminah (HSA), Johor Bahru, Hospital Sultan Ismail (HSI), Johor Bahru, Hospital Pakar Sultanah Fatimah (HPSF), Muar, Hospital Sultanah Nora Ismail (HSNI), Batu Pahat, Hospital Enche Besar Hajjah Khalsom (HEBHK), Kluang and Hospital Segamat (HDS), Segamat [17].

### Study Population

The study population consisted of adult patients aged 18 years above attending the eye clinics with a clinical diagnosis of microbial keratitis and adult in-patients aged 18 years above treated for microbial keratitis with no prior referral from the eye clinic in the selected hospital. This study did not include reports or samples or isolates originating from vitreous/aqueous, anterior chamber, orbital or washing fluid, biopsy tissue, lacrimal gland/canal, enucleation, intraocular and other non-routine specimens or reports

of patients presenting with viral or Acanthamoeba keratitis or ulcers reported due to burn or chemical exposure and minors aged 0-17 years old as the prevalence of microbial keratitis among this age group is low.

### Study Ethics

This study involved patients clinically diagnosed with bacterial keratitis who received treatment in six government hospitals offering ophthalmological services in Johor, Malaysia. The Malaysian Research Ethics Committee ethically approved this study (NMRR-19-545-45941(IIR)).

### Statistical Analysis

Data on socio-demographic factors (gender, age, ethnicity, occupation, and underlying medical illness), risk factors, and symptoms were collected from clinical notes. Statistical analysis for descriptive analysis was performed using SPSS software version 26.0 (IBM Corp., Armonk, NY).

## RESULTS

A total of eighty-nine patients were identified with bacterial keratitis from all six hospitals. Out of these 89 patients, 55 patients (61.8%) were culture positive with *Pseudomonas aeruginosa*, 21 patients (23.6%) were infected with gram-positive organisms such as *Staphylococcus aureus*, Coagulase-negative *Staphylococcus* and few others and 13 patients (14.6%) were infected with gram-negative other than *Pseudomonas aeruginosa* such as *Pseudomonas* spp., *Enterobacter* spp. and few others. The list of bacteria isolated listed in Table I.

The mean age was 41.2 ± 15.7 years (range: 18–79 years). Most patients were male (n = 36, 65.5%) and belonged to the Malay ethnicity (n = 33, 60%). This study divided the data for occupation into eight categories, namely office jobs (n = 14, 25.5%), other jobs such as night market seller, shoe vendors (n = 10, 18.2%), retirees (n = 8, 14.5%), agricultural workers such as palm oil worker (n = 7, 12.7%), students (n = 5, 9.1%), construction workers (n = 4, 7.3%), factory workers (n = 4, 7.3%), and housewives (n = 3, 5.5%). More than 70% of the patients did not have any underlying medical illness (n = 41, 74.5%), while the rest of the patients had diabetes (n = 6, 10.9%), hypertension (n = 7, 12.7%), hyperlipidemia (n = 3, 5.5%), other types of systemic diseases such as thyroid storm, cancer, and others (n = 6, 10.9%). (Table II).

In this study, five major risk factors were reported in the patients diagnosed with *Pseudomonas* keratitis in this region. The risk factors are contact-lens usage (n = 25, 45.5%), followed by ocular trauma (n = 20, 36.4%), ophthalmic steroid usage (n = 16, 29.1%), ocular surface disease (n = 12, 21.8%), and previous

**Table I : Bacteria Isolated Among Patients In Selected Government Hospitals In Johor, Malaysia**

Organism	Frequency (n)	Percentage (%)
<i>Pseudomonas aeruginosa</i>	55	61.8
<i>Staphylococcus aureus</i>	9	10.1
<i>Coagulase Negative Staph</i>	4	4.5
<i>Citrobacter koseri</i>	1	1.1
<i>Bartonella sp.</i>	1	1.1
<i>Serratia marcescens</i>	2	2.2
<i>Streptococcus pneumoniae</i>	1	1.1
<i>Acinetobacter iwoffii</i>	1	1.1
<i>Moraxella laanata</i>	1	1.1
<i>Klebsiella terrigena</i>	1	1.1
<i>Achromobacter xylosoxidans</i>	1	1.1
<i>Enterobacter cloacae</i>	1	1.1
<i>Klebsiella pneumoniae</i>	1	1.1
<i>Pseudomonas luteola</i>	1	1.1
<i>Pseudomonas stutzeri</i>	1	1.1
<i>Brevibacterium frigoritolerans</i>	1	1.1
<i>Staphylococcus epidermidis</i>	1	1.1
<i>Bacillus paramycooides</i>	1	1.1
<i>Bacillus albus</i>	1	1.1
<i>Cellulomonas flavigena</i>	1	1.1
<i>Bacillus proteolyticus</i>	1	1.1
<i>Staphylococcus saprophyticus</i>	1	1.1
<i>Pseudomonas montaiilli</i>	1	1.1
Total	89	100.0

eye surgery (n = 4, 7.3%) (Table III).

Table III lists the symptoms reported among patients with *Pseudomonas* keratitis. About 47% of the patients had three symptoms (n = 26, 47.3%) when the infection set in. More than 85% of the patients reported experiencing redness (n = 49, 89.1%) followed by eye discharge (n = 25, 45.5%), blurry vision (n = 23, 41.8%), itchiness (n = 5, 9.1%), and other symptoms such as grittiness and sensitivity (n = 28, 50.9%) during the onset of the infection (Table IV).

Further analysis was conducted using Chi-square to find the association between socio-demographic factors, risk factors and symptoms with *Pseudomonas* keratitis. The analysis found a significant association between underlying medical illness (p=0.036), and ocular surface disease (p=0.051), The rest of the factors are not significantly related with *Pseudomonas* keratitis (Table V).

**Table II : Sociodemographic Data Of Patients With *Pseudomonas* Keratitis**

Demographic variable	Frequency (n)	Percentage (%)
<b>Age</b>		
< 60	45	81.8
> 60	10	18.2
<b>Gender</b>		
Male	36	65.5
Female	19	34.5
<b>Ethnicity</b>		
Malay	33	60.0
Chinese	9	16.4
Foreigner	7	12.7
Indian	6	10.9
<b>Occupation</b>		
Office	14	25.5
Other jobs	10	18.2
Retired	8	14.5
Agriculture	7	12.7
Student	5	9.1
Construction	4	7.3
Factory	4	7.3
Housewife	3	5.5
<b>Underlying Medical Illness*</b>		
No known medical illnesses	41	74.5
Diabetes	6	10.9
Hypertension	7	12.7
Hyperlipidemia	3	5.5
Others	6	10.9

\*some patients had more than one underlying medical illness

**Table III : Predisposing Risk Factors For *Pseudomonas* Keratitis**

Predisposing risk factor*	Frequency (n)	Percentage (%)
Contact lens	25	45.5
Ocular trauma	20	36.4
Ophthalmic steroid usage	16	29.1
Ocular surface disease	12	21.8
Previous eye surgery	4	7.3

\*some patients had more than one risk factor

## DISCUSSION

*Pseudomonas aeruginosa* was first isolated and named *Bacillus pyocyaneus* in 1882. Later, in 1891, Sattler

**Table IV : Reported Symptoms For *Pseudomonas* Keratitis**

Symptom	Frequency (n)	Percentage (%)
<b>Number of symptoms</b>		
One	6	10.9
Two	23	41.8
Three	26	47.3
<b>Type of symptom</b>		
Redness	49	89.1
Discharge	25	45.5
Blurry vision	23	41.8
Itchiness	5	9.1
Others	28	50.9

reported the first case of corneal ulcer caused by *Bacillus pyocyaneus* i.e., *Pseudomonas aeruginosa*. Since then, *Pseudomonas aeruginosa* has become the most dominant pathogen in microbial keratitis [18]. Keratitis caused by *Pseudomonas aeruginosa* often progresses rapidly, with onset occurring within 24 hours after the bacterium comes in contact with the corneal layer. This kind of keratitis usually sets in when there is trauma to the corneal layer, or there is a contact with a contaminated soft lens or contact-lens cleaning solution, and also among immunocompromised individuals [19].

Although *Pseudomonas aeruginosa* often addressed as an opportunistic pathogen causing infection only in immunocompromised individuals [20], yet it has 18 different types of virulence effectors which are important components in causing infection in mammals [6]. Among these virulence factors, ExoU regarded as the most toxic effector protein which can cause severe disease with poor prognosis due to the fast and irreversible damage secreted by the phospholipase activity of ExoU [6]. On the other hand, Callaghan et al reported protease IV (PIV) and *Pseudomonas aeruginosa* small protease (PASP) as the main endotoxins isolated from strains of *Pseudomonas aeruginosa* that causes corneal ulcer or keratitis. PIV is able to destruct host defensive proteins and induce inflammation while PASP is capable of causing erosion on collagen-rich cornea. These exclusively *Pseudomonas*-secreted proteases are the major virulence factors in *Pseudomonas* keratitis [21].

In the present study, *Pseudomonas* keratitis showed a preponderance of males more than females. This pattern of gender is similar in all Malaysian studies [8,22,23] and many studies globally, especially from the Asian continent [2,24,25]. The preponderance exists due to differences in the risk factors that lead to keratitis. In Asia, males are predominantly involved in activities risking eye injury such as outdoor activities,

**Table V : Association Between Various Factors and *Pseudomonas* Keratitis**

Factors	Frequency (n)	$\rho$ Value
<b>Socio-demographic</b>		
<b>Age</b>		
< 60	45	0.542
> 60	10	
<b>Gender</b>		
Male	36	0.724
Female	19	
<b>Ethnicity</b>		
Malaysian	48	0.298
Non-Malaysian	7	
<b>Occupation</b>		
Office	14	0.599
Other jobs	10	0.779
Retired	8	0.284
Agriculture	7	0.298
Students	5	0.233
Construction	4	0.108
Factory	4	0.792
Housewife	3	0.578
<b>Underlying Medical Illness</b>		
No known medical illnesses	41	<b>0.036*</b>
Having medical illness	14	
<b>Risk Factors</b>		
Contact lens	25	0.290
Ocular trauma	20	0.919
Ophthalmic steroid usage	16	<b>0.057*</b>
Ocular surface disease	12	<b>0.051*</b>
Previous eye surgery	4	0.792
<b>Symptoms</b>		
Redness	49	<b>0.057*</b>
Discharge	25	0.205
Blurry vision	23	0.628
Itchiness	5	0.415
Others	28	0.649

\* $p < 0.05$ = statistically significant, ( $\rho$ = Chi square)

handling heavy machineries, working on high-rise buildings and many other risky activities compared to females [26]. Nevertheless, 81.8% of the patients in this study are below 60 years old, which falls into the productive age group. This productive age group are actively involved in the workforce in this region. Again, a similar trend was observed in most of the Malaysian studies [27,28] and in most studies from the

Asian continent [29–31]. As for ethnicity, the pattern is also similar to almost all of the studies in Malaysia, as Malays are the major ethnic in the country, making up almost 60% of the population in this region [32].

A quarter ( $n = 14$ , 25.5%) of the cases were reported among the patients involved in office-oriented jobs, namely office workers such as management officers, clerks, teachers, insurance agents and many more. According to Department of Statistics Malaysia census in 2022, more than half of the labour force in Johor works in the service sector (59.5%), followed by the manufacturing sector (24.5%), construction (8.4%), and agriculture (7.2%) [32]. Besides, these office workers are the most active users of contact lens; in this study, 45.5% of the cases presented with this risk factor. This finding is in line with other studies in Malaysia, which reported contact-lens usage as the main risk for keratitis caused by *Pseudomonas aeruginosa* [8,33]. Its unique structure consists of flagella, pili, lipopolysaccharide, and extracellular products, including proteases, exotoxin A gives the advantage for *Pseudomonas aeruginosa* to adhere and form biofilm on any surface especially contact-lens. In fact, the strains found on contact lens surfaces contains more ExoU-positive proteins compare to strains found on non-contact lens material [34].

This study also showed that those infected with *Pseudomonas aeruginosa* reported redness, eye discharge, and blurry vision as common symptoms during the onset of the infection. These symptoms are common in bacterial keratitis, especially *Pseudomonas aeruginosa*, as the organism produces enzymes such as protease and elastase and toxins such as exotoxin A, which causes rapid lysis of the tissues on the corneal surface, manifested by the above symptoms [34].

In this study, the analysis of association between all socio-demographic factors, risk factors and symptoms and *Pseudomonas aeruginosa* showed a significant association with two factors listed earlier. The socio-demographic factor that was significantly associated with *Pseudomonas* keratitis was underlying medical illness ( $p=0.036$ ). This association indicated although the incidence of underlying medical illness among cases in this region low, yet having at least having one underlying medical illness can lead to poor infection prognosis. Among the diseases, diabetes is one of the most commonly occurring non-communicable diseases in Malaysia. The National Health and Morbidity Survey 2019 stated that the prevalence of diabetes among Malaysians was 18.1%, and the same survey revealed that 1 in 5 adults in Malaysia above 18 years old has diabetes [35]. Diabetes mellitus has an extreme sequel on multiple organ systems and the most affected organ is the eyes. Diabetes negatively affects the dynamics of the blood-retinal barrier, which modifies regular

cell-cell interactions and leads to profound vascular abnormalities, loss of the blood-retinal barrier and impaired neuronal function [36].

Another factor found to be significantly associated with *Pseudomonas* keratitis was ocular surface disease ( $p=0.051$ ). The eyes are a tiny complex organ among all organs in the human body and any disease, be it microbial keratitis or ocular surface disease, will be complex and challenging to treat and manage. According to the Cornea, External Disease, and Refractive Society (CEDARS), the complexity of the ocular surface disease is divided into five subtypes; Aqueous deficiency, blepharitis/meibomian gland dysfunction (evaporative and non-evaporative), goblet cell deficiency/mucin deficiency, exposure (unable to close eyes completely) and dysfunctional tear syndrome/co-conspirators [37]. In *Pseudomonas* keratitis, ocular surface disease, an existing altered condition of the cornea, becomes favourable for the organism to further the damage by rapidly producing the proteases to lysis and breakdown the collagen membrane of the cornea thus causing more severe infection and in the end even more difficult to treat [38,39].

## CONCLUSION

*Pseudomonas* keratitis is a fast and devastating infection that requires fast and effective treatment. In this study, like other studies on keratitis, males are affected more than females. Contact lens usage found in majority of cases in the present study as *Pseudomonas aeruginosa* is commonly found to adhere to the contact lens surface via the formation of biofilm. However, risk factors ocular surface disease was significantly associated with *Pseudomonas* keratitis. The present study is the first in Malaysia which specifically reported on *pseudomonas* keratitis and its epidemiology. The data from this study will serve as a baseline data for industrial and healthcare policy makers on designing eye care awareness-related activities and eye injury preventive measures. Future studies include extending this study to other parts of Malaysia and investigating.

## ACKNOWLEDGEMENTS

Thank you to all data providers from Hospital Sultanah Aminah and Hospital Sultan Ismail from Johor Bahru district, Hospital Pakar Sultanah Fatimah from Muar district, Hospital Sultanah Nora Ismail from Batu Pahat district, Hospital Segamat from Segamat district and Hospital Enche Besar Hajjah Khalsom from Kluang district. This manuscript is supported by Universiti Putra Malaysia initiative grant (GP-IPS 2019/9680700).

## REFERENCES

1. Ouattara Y, Diabaté Z, Godé L, et al.: Epidemiological, Clinical, Therapeutic and

- Evolutionary Aspects of Corneal Abscesses at the Teaching Hospital of Bouaké (Côte d'Ivoire). *Ophthalmol Res.* 2021; 4:6. doi: 10.33425/2639-9482.1019
2. Dong PN, Hang DTT, Duong NTN, Lien MT, Chen AC, Aldave AJ: Infectious keratitis in Vietnam: etiology, organisms, and management at Vietnam National Eye Hospital. *Int J Ophthalmol.* 2022; 15:128–34. doi:10.18240/ijo.2022.01.19
  3. Marquart ME, O'Callaghan RJ: Infectious Keratitis: Secreted Bacterial Proteins That Mediate Corneal Damage. *J Ophthalmol.* 2013; 2013:1–9. doi:10.1155/2013/369094
  4. Sy A, Srinivasan M, Mascarenhas J, et al.: *Pseudomonas aeruginosa* Keratitis: Outcomes and Response to Corticosteroid Treatment. *Investig Ophthalmology Vis Sci.* 2012; 53:267. doi:10.1167/iovs.11-7840
  5. Fleiszig SM, Evans DJ: The pathogenesis of bacterial keratitis: studies with *Pseudomonas aeruginosa*. *Clin Exp Optom.* 2002; 85:271–8. doi:10.1111/j.1444-0938.2002.tb03082.x
  6. Foulkes DM, McLean K, Haneef AS, Fernig DG, Winstanley C, Berry N, Kaye SB: *Pseudomonas aeruginosa* Toxin ExoU as a Therapeutic Target in the Treatment of Bacterial Infections. *Microorganisms.* 2019; 7:707. doi: 10.3390/microorganisms7120707
  7. Oka N, Suzuki T, Ishikawa E, Yamaguchi S, Hayashi N, Gotoh N, Ohashi Y: Relationship of Virulence Factors and Clinical Features in Keratitis Caused by *Pseudomonas aeruginosa*. *Investig Ophthalmology Vis Sci.* 2015; 56:6892. doi: 10.1167/iovs.15-17556
  8. Hooi SH, Hooi ST: Culture-Proven Bacterial Keratitis in a Malaysian General Hospital. *Med J Malays.* 2005; 60:614–23.
  9. Lim Wen Siang J, Wu Zhuan O, Yi Chen N, Sok Lin N. Profile of Microbial Keratitis. *Cureus.* 2021;13(12):e20663. doi:10.7759/cureus.20663
  10. Tananuvat N, Punyakhum O, Ausayakhun S, Chaidaroon W. Etiology and clinical outcomes of microbial keratitis at a tertiary eye-care center in northern Thailand. *J Med Assoc Thai.* 2012; 95:S8-17.
  11. Al-Shakarchi FI, Hussein MA, Department of Biotechnology, College of Science, Al-Nahrain University., Al-Shaibani AB, Department of Biotechnology, College of Science, Al-Nahrain University.: Profile of Microbial Keratitis at a Referral Center in Iraq. *J Al-Nahrain Univ-Sci.* 2015; 18:141–7. doi:10.22401/JNUS.18.1.21
  12. Ng AL-K, To KK-W, Choi CC-L, et al.: Predisposing Factors, Microbial Characteristics, and Clinical Outcome of Microbial Keratitis in a Tertiary Centre in Hong Kong: A 10-Year Experience. *J Ophthalmol.* 2015, 2015:1–9. doi:10.1155/2015/769436
  13. Rizwan A, Asghar A, Naqvi SAH, Sughra U, Raza H: Risk factors, causative organisms and sensitivity patterns of infective keratitis in a tertiary care hospital in Rawalpindi. *J Pak Med Assoc.* 2021; 71:5. doi: 10.47391/JPMA.1410
  14. Tong W, Chen D, Chai C, Tan AM, Manotosh R: Disease patterns of microbial keratitis in Singapore: A retrospective case series. *Contact Lens Anterior Eye.* 2019; 42:455–61. doi: 10.1016/j.clae.2019.02.006
  15. Vadivoo NS, Sujatha K, Sridevi NVK, Sharda R, Niranjana M: Ten Year Study of Fungal Keratitis-Tertiary Care Hospital. *Natl J Basic Med Sci.* 2012; 3:149–53. doi:10.13140/RG.2.2.24146.66241.
  16. Johor (State, Malaysia) - Population Statistics, Charts, Map and Location. Accessed: November 16, 2021. [https://www.citypopulation.de/en/malaysia/admin/01\\_johor/](https://www.citypopulation.de/en/malaysia/admin/01_johor/).
  17. Portal Rasmi Kementerian Kesihatan Malaysia. Accessed: March 7, 2021. [https://www.moh.gov.my/index.php/database\\_stores/store\\_view/3?search=johor](https://www.moh.gov.my/index.php/database_stores/store_view/3?search=johor).
  18. Hyndiuk RA. Experimental *pseudomonas* keratitis. *Trans Am Ophthalmol Soc.* 1981; 79:540.
  19. Rosenkranz L: *Pseudomonas* Keratitis. *Am J Ophthalmol.* 1983; 95:566–7. doi:10.1016/0002-9394(83)90292-1
  20. Hilliam Y, Kaye S, Winstanley C: *Pseudomonas aeruginosa* and microbial keratitis. *J Med Microbiol.* 2020, 69:3–13. doi:10.1099/jmm.0.001110
  21. O'Callaghan R, Caballero A, Tang A, Bierdeman M: *Pseudomonas aeruginosa* Keratitis: Protease IV and PASP as Corneal Virulence Mediators. *Microorganisms.* 2019; 7:281. doi:10.3390/microorganisms7090281
  22. Omar N, Muna'aim MA, Saleh RM, Kasim ZM, Isa MM: An 8-year Retrospective Review of Microbial Keratitis in A Secondary Referral Centre in Malaysia. *Malays J Med Health Sci.* 2017; 13:47–57.
  23. Bou KJ, Siew TT, Wei CT, Mun TL. Infectious keratitis: findings from a retrospective review in the central zone of Sarawak. *Malays J Ophthalmol.* 2019; 3:181–96. doi:10.35119/myjo.v1i3.61
  24. Reza MS, Chowdhury SA, Talukder PK, Islam MT, Hossain MA, Hussain NJ, Jhuma AA: Non-viral Microbial Keratitis in a Tertiary Care Hospital in Bangladesh. *Med Today.* 2021; 33:133–7. doi:10.3329/medtoday.v33i2.56059
  25. Khan N, Banerjee G, Gupta P, Kishore P, Sharma A: Etiological diagnosis of microbial keratitis in a tertiary care hospital in Lucknow. *Kerala J Ophthalmol.* 2021; 33:39. doi:10.4103/kjo.kjo\_85\_20
  26. Ting DSJ, Ho CS, Cairns J, et al.: 12-year analysis of incidence, microbiological profiles and in vitro antimicrobial susceptibility of infectious keratitis: the Nottingham Infectious Keratitis Study. *Br J Ophthalmol.* 2021; 105:328–33. doi:10.1136/bjophthalmol-2020-316128
  27. Khor HG, Cho I, Lee KRCK, Chieng LL. Spectrum of Microbial Keratitis Encountered in the Tropics:

- Eye Contact Lens Sci Clin Pract. 2020; 46:17–23. doi:10.1097/ICL.0000000000000621
28. Ratnalingam V, Umapathy T, Sumugam K, Hanafi H, Retnasabapathy S. Microbial keratitis in West and East Malaysia. *Int Eye Sci.* 2017; 17:1989–92. doi:10.3980/j.issn.1672-5123.2017.11.01
  29. Hoffman JJ, Yadav R, Sanyam SD, et al. Microbial Keratitis in Nepal: Predicting the Microbial Aetiology from Clinical Features. *J Fungi.* 2022; 8:201. doi: 10.3390/jof8020201
  30. Bajracharya L, Bade AR, Gurung R, Dhakhwa K: Demography, Risk Factors, and Clinical and Microbiological Features of Microbial Keratitis at a Tertiary Eye Hospital in Nepal. *Clin Ophthalmol.* 2020; 14:3219–26. doi:10.2147/OPTH.S266218
  31. Shahi R, Sareen D, Maurya OPS, Tilak R, Prajapati M: Clinical and microbiological spectrum of corneal ulcer in eastern region of Uttar Pradesh. *Indian J Clin Exp Ophthalmol.* 2019; 5:182–5. doi:10.18231/j.ijceo.2019.043
  32. Mahidin DSDMU: POCKET STATS Q3 2021. DEPARTMENT OF STATISTICS MALAYSIA; 2021.
  33. Kadir AJ, Samsudin A, Fauzi A, Abidin ZZ. Review of corneal ulcers at University Malaya Medical Centre. *Int J Ophthalmol.* 2008; 8:2376–80.
  34. Suzuki T, Inoue H: Mechanisms Underlying Contact Lens-Related Keratitis Caused by *Pseudomonas aeruginosa*. *Eye Contact Lens Sci Clin Pract.* 2022; 48:134–7. doi:10.1097/ICL.0000000000000880
  35. Institute for Public Health: National Health and Morbidity Survey (NHMS) 2019 (NMRR-18-3085-44207) Non-communicable diseases, healthcare demand, and health literacy: Key Findings. 2020.
  36. Antonetti DA, Silva PS, Stitt AW: Current understanding of the molecular and cellular pathology of diabetic retinopathy. *Nat Rev Endocrinol.* 2021; 17:195–206. doi:10.1038/s41574-020-00451-4
  37. McKinney S: Diagnosing Ocular Surface Disease. *Rev. Ophthalmol.* (2019). Accessed: December 7, 2021. <https://www.reviewofophthalmology.com/article/diagnosing-ocular-surface-disease>.
  38. Green M, Apel A, Stapleton F: Risk Factors and Causative Organisms in Microbial Keratitis. *Cornea.* 2008; 27:22–7. doi:10.1097/ICO.0b013e318156caf2
  39. Subedi D, Vijay AK, Willcox M: Overview of mechanisms of antibiotic resistance in *Pseudomonas aeruginosa*: an ocular perspective. *Clin Exp Optom.* 2018; 101:162–71. doi: 10.1111/cxo.12621