

Seasonal Presentation and Microbiological Profile of Infectious Keratitis (IK) in Selected Hospitals in Ilorin, Nigeria.

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Abstract

Background: Infectious keratitis is a common ophthalmic disease with the potential for severe ocular morbidity. Although, there have been few reports of ocular infections from other geopolitical regions in Nigeria but Seasonal variation in patients' presentation to hospital is yet to be described in this centre. This study sets out to determine the seasonal trends of onset of infectious keratitis and causative microorganisms in selected hospitals in Ilorin, Nigeria. **Methods:** This was a cross-sectional study of patients who presented with infectious keratitis at the Ophthalmology Clinics of selected hospitals in Ilorin. Seasonal distribution of incidence, demographic characteristics, culture and microbiological profiles of the patients were determined. We analyzed the data using SPSS version 20.0 software to determine whether a significant variation existed between rainy and seasons, frequency of presentation of ulcers and other risk factors. **RESULTS:** A total of 89 patients; 69 Male (34 in rainy season, 25 in dry season) and 30 Female (20 in rainy season and 10 in dry season), with a mean age of 40 (range 3-77; median, 36) diagnosed with infectious keratitis was included in the analysis. Bacterial culture positivity rate for the rainy and dry season were 36 (59.0%), 25 (40.9%) respectively. Significant variations observed in causative organisms; *Staphylococcus species 14* (22.9%) in rainy season, *Klebsiella 6*(9.8%) in dry season. **CONCLUSION:** Incidence of bacterial-induced infections was high in rainy season. The association of *staphylococcus aureus* and *pseudomonas aeruginosa* infection with rainy season was likely attributed to warmer temperature conducive for microbial growth.

Keywords: Seasonal variation; Corneal infection; Infectious keratitis; Staphylococcus; Pseudomonas.

INTRODUCTION

Infectious keratitis (IK) is the leading cause of corneal blindness in both developed and developing countries,^{1,2} and is responsible for approximately 2 million monocular blindness per year, with higher rates reported in developing countries.^{3,4,5} A wide range of microorganisms, including bacteria, fungi, viruses and parasites have been implicated.^{6,7}

Seasonal variations occur periodically in a year. Geographical and temporal variations of IK have been well reported in the literature^{8,9,10}, with bacteria and

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How to cite this article: Oladejo *et al*: Seasonal Presentation and Microbiological Profile of Infectious Keratitis (IK) in Selected Hospitals in Ilorin, Nigeria. *Ann Trop Pathol.*, 2024; 15 (2): 42-46

fungi being the most common microorganisms responsible for IK in developed and developing countries respectively.^{3,9,10} The study aimed to investigate the seasonality of infectious keratitis by pathogens.

MATERIALS AND METHODS

Study Setting

This study was conducted at the Ophthalmology Clinic of University of Ilorin Teaching Hospital (UITH), Sobi Specialist Hospital and Civil Service Clinic which serve as referral hospitals in Kwara State, North Central, Nigeria, between July 2015 and July 2018. Ethical permission for the study was obtained from Ministry of Health, Ilorin, Kwara State with approval code: MOH/KS/EC/777/88/24. In Nigeria, most significant rainfall occurs from April to October while dry season (harmattan period) is from November to March.

Study Design

This is a cross-sectional descriptive study of patients with infectious keratitis who attended Ophthalmology Clinic, University of Ilorin Teaching Hospital, Sobi Specialist Eye Clinic and Civil Service Eye Clinic in Ilorin which serve as referral centers for ophthalmic diseases in Kwara State, Nigeria, between July 2015 and July 2018.

Study Participants and Selection Criteria

A complete history was taken from each participant about demographic characteristics such as age, sex and occupation. Eyes with clinically suspected viral and parasitic corneal ulcers, previous corneal disease, use of antimicrobial medication immunosuppressive were excluded. History of pain, photophobia, watering, and redness as well as predisposing factors like trauma, contact lens wear, dry eye and surgery was taken. Seasonal incidence among the dominant rainy and dry seasons of the year were noted. The ocular examination included visual acuity (VA) of both eyes taken independently with Snellen's chart at distance of 6 meters from the patient position, and slit lamp examination of the cornea for size, site, and depth of the ulcer, presence or absence of perforation. Fluorescein staining of the corneal ulcer for epithelial defect measurements and the presence or absence of hypopyon was also determined.

Cornea Sample Collection

Informed consent obtained after explaining the procedure to the patient, non-preservative topical anaesthesia instilled into the infected eye. The patient was thereafter positioned at the Slit Lamp Biomicroscope, the corneal scraping was done by the Ophthalmologist. Immediately after collection, the samples were inoculated directly into brain heart infusion broth (BHB), yeast extract broth (YEB), and Tris EDTA buffer which was stored at temperature of -80°C. Smears of the samples were made on the slide for Gram staining and the remaining sample was streaked directly on Blood agar, Chocolate and MacConkey agar. The culture samples thereafter were transported to the Medical and Parasitology Laboratory in University of Ilorin Teaching Hospital where culture isolation and identification of Gram-negative, Gram-positive organisms, fungal pathogens especially aspergillus, fusarium species and candida species were done.

Gram stain for cellular morphology was done to characterize each isolate.¹³ Further characterization was done by subjecting colonies to biochemical tests and Analytical profile index (API) to identify the bacteria to species. The resulting reactions were read according to the reading table attached with the API kit and the identification was obtained by referring to the identification software on Apiweb (<http://apiweb.biomerieux.com>).

The yeast extract broth (YEB) was cultured onto Sabouraud dextrose agar (SDA) and were incubated at 28°C for five days. Lactophenol cotton blue wet mount preparation was carried out on the growth on Sabouraud dextrose agar (SDA) plates while Mycology Atlas and biochemical tests were used for the identification of the fungi. Suspected yeast isolates from Sabouraud dextrose agar (oxid, UK) were Gram's stained and confirmed yeast isolates were further characterized according to Matare *et al*.¹⁴

Statistical Analysis

The data were entered into and analyzed using the Statistical Package for Social Scientist (SPSS) version, Chicago 20.0, USA software. Percentages, mean and standard deviation was used to describe numerical variables. Student 't' test and analysis of variance (ANOVA) was used to compare means of two or more independent variables respectively. Independent categorical variables were compared using the Chi squared test. Confidence interval was set at 95% and for all statistical test $p < 0.05$ was considered significant. Presentation was done with the use of frequency tables, charts and figures.

RESULTS

The baseline characteristics of the participants (89 patients), age group ranged from <20 years, 21-49 years and >50 years with a male to female ratio 3:1 is shown in table 1 below. There were more patients in rainy season 54(60.7%) than dry season 35(39.3%) while farmers were more predisposed to infectious keratitis 35(39.3%). Table 1: Age Range, Gender, Occupation and Seasonal Distributions of Infectious Keratitis

Age range/ years	Gender and distributions				
	Rainy season		Dry season		Total
	M	F	M	F	
< 20	3	2	2	1	8(8.9%)
21-49	23	12	16	7	58(65.2%)
>50	8	6	7	2	23(25.8%)
Total	34	20	25	10	89(100.0%)
Occupation	Rainy season		Dry season		Total
Artisan	8	5	2	5	
Farmer	10	13	7	5	35(39.3%)
Civil servant	3	1	2	1	7(7.9%)
Trader	8	3	2	4	17(19.1%)
Students	2	2	2	1	7(7.9%)
Others	2	0	1	0	3(3.4%)
Total	33	24	16	16	89

M = Male; F = Female

Table 2: Corneal Ulcer Size, Its Location and Seasonal Distribution

Corneal ulcer size/ mm	Ulcer location & Seasonal distribution				
	Central		Paracentral		Total
	Rain	Dry	Rain	Dry	
1-2	27	13	10	7	57(64.0%)
3-4	16	5	5	2	28(31.4%)
>4	2	1	1	-	4(4.5%)
Total	45	19	16	9	89

P<0.001

The mean duration of patients presenting to hospital was 7 days. Many patients had symptoms for 7-10 days.

Centrally located corneal ulcers were evident in many patients 64(71.9%). Majority of the ulcers 57(64.0%) measure 1-2mm in size while 4(4.5%) of the patients had ulcer size > 4mm as stated in table 2.

The culture positivity rate in rainy season was higher than dry season. Out of 89 corneal scraping samples, 61 bacterial isolates were culture positive while 18 fungal isolates were culture positive. Ten corneal samples were culture negative as shown in table 3.

Table 3: Seasonal Variation of Isolates Using API and Conventional Methods

Isolates	No	Dry season	Rain season	Total
Culture results				
Positive		25(40.9%)	36(59.0%)	61
Negative		6	4	
Staphylococcus		8(13.1%)	14(22.9%)	P<0.005
Pseudomonas		3(4.9%)	9(14.8%)	P<0.005
Klebsiella		6(9.8%)	2(3.3%)	
Other Gram-ve bacilli		4(6.6%)	7(11.5%)	
Uncommon pathogens		4(6.6%)	4(.6%)	
Fungi				
Culture results				
Positive		6(33.3%)	12(66.7%)	18
Candida albican		1(5.6%)	4(22.2%)	P<0.019
Trichophyton rubrum		-	2(11.1%)	
Fusarium solani		4(22.2%)	3(16.7%)	
Aspergillus fumigatus		1(5.6%)	3(16.7%)	

DISCUSSION

Seasonal variation is a common feature of infectious keratitis.¹⁵ Although both bacteria and fungi are known to cause infectious keratitis, the etiological agents vary from region to region depending upon geographical area which is also depending on environmental conditions.¹⁶

In this study, the incidence of infectious keratitis was higher among males in rainy season than dry season which is similar to the study conducted by Saka *et al*¹⁷ in Ilorin, Tewari *et al* in India,¹⁸ Ibanga *et al*¹⁹ and Kieran *et al*.²⁰ The gender variation in incidence of infectious keratitis might be due to variation in geographical locations However, the females were more predisposed to occupation-induced corneal ulcers especially farming

in rainy season in this study. The gender variation in incidence of infectious keratitis might also be due to variation in geographical locations. This is similar to the study done in China that reported women as predominantly affected by gender.²¹ Tan et al in United Kingdom,²² Lap-King et al in China²³ and Gorski et al in United Kingdom²⁴ reported high number of females. These are also attributed to agricultural, domestic workers and laborers. In this study, corneal infection was seen in all age groups with preponderance among the young and young adult age group in rainy season. This is different from the study conducted by Saka et al¹⁷ and Pei et al²⁵ that showed adult as the predominant age group affected by infectious keratitis. This variation might be due to geographical locations and their age range differences among farmers, domestic workers and laborers, most cases residents are rural dwellers.

There was a higher incidence of keratitis among farmers in this study in rainy season than dry season. This is similar to the studies conducted by Tewari et al¹⁸ and Suwal et al²⁶ but is different from Ranjini et al²⁷ study that reported a higher incidence of keratitis among housewives.

This study showed that agricultural workers were affected more than the general population in rainy season. This is attributed to the fact that those working in agricultural fields are more prone to vegetative trauma especially in planting season period. In this study, a maximum number of cases and culture positivity were seen in rainy season which is characterized by warm and humid period but is different from Nin et al²⁸ who reported a maximum number of cases during the dry season which happens to be harvesting months in South India. This may be due to geographical variation in planting seasons.

This study showed that more than seventy percent of patients had centrally located corneal ulcer which was similar to the findings of Saka¹⁷ in Ilorin. The rainy season had higher frequency of infectious keratitis than the dry season. The finding is higher compare to the study done by Gorski²⁴ who reported lower frequency of infectious keratitis in rainy season. The higher value in this study may be due to smaller sample size and different geographical location.

There was higher rate of culture positivity in the rainy season than dry season in this study. The higher culture positivity in this study is different from the findings by Walkden et al¹² and Gorski et al²⁴ that reported higher culture positivity during dry season compare to rainy season. This may be due to seasonal variation in different geographical regions.

In this study, significant seasonal variations in *Staphylococcus species* and *Klebsiella species* was seen. *Staphylococcus aureus* and *Pseudomonas aeruginosa*

was commonly observed during rainy season compare to dry season. This is similar to what was reported in other studies.^{19,24}

The fungal keratitis during the rainy season was higher than the dry season in the present study. This is similar to the study by Ung et al who demonstrated a significantly higher rate of fungal infection during rainy season in Southeast India.³⁰ Such variations have been linked to environmental factors, such as humidity, rainfall, wind, and also to the harvest periods. Several studies from other countries have reported about infectious keratitis and its seasonality.¹²

CONCLUSION

The rainy season has higher frequency of infectious keratitis and *Pseudomonas aeruginosa*. Possible factors leading to this increased rainy season incidence include warmer temperatures and higher humidity. Knowledge of seasonal variations in pathogenic organisms will help Clinicians, including Ophthalmologists and non-ophthalmologists, who work at the front-line service such as accident and emergency department and primary care setting in the choice of antimicrobial agents in clinical practice.

Funding Acquisition: None

Conflict of interest: None

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