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ORIGINAL RESEARCH

Assessment of the Causative Agents, Systemic Risk Factors, and Antimicrobial Sensitivity Of Keratitis Infection Among Patients Admitted at Madhubani Medical College And Hospital, Madhubani, Bihar

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ABSTRACT

Aim: Assess the Causative agents, Systemic Risk Factors, and antimicrobial Sensitivity of Keratitis Infection Among Patients Admitted at Madhubani medical college and hospital, Madhubani, Bihar. **Material and methods:** A quantitative, non-experimental research design is an analytical cross-sectional study. This type of research aims to "collect data from a group of subjects at only one point in time. A total of 400 people met the inclusion criteria, and a researcher was selected to conduct the primary study form January 2023 to December 2023. In the end, 400 patients were present throughout the full data collection process. Patients of all sexes and ages who have been diagnosed with infectious keratitis are included in this category. Demographic data like Age, Gender, Occupation, Area of residency, Co morbid illness associated with Keratitis, Medications used, Trauma factors. The methods used for Specimen processing wet mount, Potassium hydroxide Method, Grams staining, Culture and Sensitivity testing were performed. **Results:** 35% of the patients were not using any medication, while 4.5% were using Corticosteroids. 80.5% of the patients associated risk factor were trauma, while 4. % were associated with the use of contact lens. 80.5% of the were infected with fungi andonly3% clients were infected with virus. It shows that 79% of the patients were positive in KOH Method , while 21 % were negative. Majority 24.68% of the Isolated Fungal species are A. fumigatus. and the least isolated one was 1.26%Fonsaceapederosoi. Table-6 It shows that 13.25 % of the patients sample reveals the presence of bacteria , while 86.75 % were negative. Majority56.60% of the gram+Ve bacteria are S. aureus. and the least isolated one was 1.88% from gram -Ve. ie A. baumannii. It shows that wet mount 3.25% (13) was positive among participants , and none of them in culture positive. Majority83.33% are Herpes simplex virus. and the least isolated one was 16.67% from Varicella zoster virus. 19.30% of the sample were resistance to Natamycinc But all samples were susceptible to ketoconazole. S.aureus isolates MSSA and MRSA 50%.It also reveals that MRSA AND MSSA isolates linezolid 100%and in MSSA , Chloramphenicol and penicillin were 100% resistant. P. aeruginosa isolates Aztreonam and Meropenem 100%. E. coli isolates all antibiotics except Ampicillin and cotrimoxazole shows 100% resistance. K. pneumoniae susceptible to three medicines, they are Imipenem, Ciprofloxacine and meropenem and all other medicines shows 100% resistance. K. pneumoniae susceptible to all medicines, except cotrimoxazole and ampicillin shows 100% resistance. **Conclusion:** Fungal corneal ulcers were found to be more common than bacterial corneal ulcers in this study. Because ofour country's tropical environment, which is both warm and humid, this is understandable. Simple bedside diagnostics, such as direct Gram staining or KOH mount and PCR test can provide valuable information on the type of the infectious agent and can assist doctors in selecting the most appropriate antibiotic treatment.

INTRODUCTION

There are three types of eye infections: those that affect the eyeball, those that affect the choroid, and those that affect the retina. Infections of the sclera include inflammatory conjunctivitis, blepharitis, keratitis, scleritis, and dacrocystitis, which are all diseases of the eyelids. A conjunctivitis that is caused by an infection is the most common type of conjunctivitis that people get. A disease of the choroidal or retinal tissues is called uveitis, while a disease of the cornea is called retinitis. It is possible that these infections are caused by outside organisms

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that get into the eye during eye trauma or surgery, or by inside organisms that have spread through the body. It is called keratitis, which is inflammation of the cornea caused by infectious organisms or other agents or stimuli. This is the most serious of the diseases we talked about. Keratitis is thought to be the most dangerous because it can cause a lot of bad things, like blindness. If you don't treat it, it can even get so bad that it can cause endophthalmitis.[1-4]

Microbial keratitis (MK) is a group of ocular infections that affect the cornea and can be caused by bacteria, fungal, and protozoal organisms. These organisms can cause ocular morbidity and disability. An individual's level of pathogenicity, his or her corneal condition, and his or her immune system all play a role in how severe a corneal infection is. There are many ways to fight off infection, like blinking, tight junctions in the corneal epithelial cells, and chemicals that stop bacteria from growing. An injury to the eye or an epithelial defect can weaken the body's defences against pathogens, which leads to infections, inflammation, and eventually blindness. Trauma, systemic diseases like diabetes and long-term use of topical corticosteroids, the use of contact lenses (wearing them overnight or for a longtime), ocular surgery (corneal surgery), inadequate disinfecting solutions, and chronic ocular surface disease are some of the most common risk factors. 120,000 people who are corneal blind live in the country right now, according to the National Programme for Control of Blindness (NPCB). It's thought that about 25,000 to 30,000 cases of corneal blindness are added each year in the country by this estimate. The fact that 90% of the world's cases of ocular trauma and corneal ulceration that lead to corneal blindness happen in developing countries shows how much of a problem corneal disease is in our country.[5-7]

MATERIAL AND METHODS

A quantitative, non-experimental research design is an analytical cross-sectional study. This type of research aims to "collect data from a group of subjects at only one point in time." It is the goal of this study to examine the link between an exposure and an illness, condition, or outcome in a given population. Surveys and questionnaires are frequently used in cross-sectional research to collect data from participants. The researcher originally intended to collect 400 samples for the study from January 2023 to December 2023, but during the recruitment stage, the present study included keratitis patients who were hospitalised and attended the outpatient departments of selected hospitals in Madhubani medical college and hospital, Madhubani, Bihar. All of the participants in the study gave their informed consent before taking part in the trial. A total of 400 people met the inclusion criteria, and a researcher was selected to conduct the primary study. In the end, 400 patients were present throughout the full data collection process.Patients of all sexes and ages who have been diagnosed with infectious keratitis are included in this category. The research will cover both in-patients and out- patients. Student who was present at the time of data collection. Student who was willing to participate were included in this study. Patients with Malignancy, Geneticmal formation, Acute illness, Pregnancy, Other valvular dysfunctions and Other Endocrine abnormalities were excluded from the study.

METHODOLOGY

The sampling procedure assists in the creation of a sample that reflects the characteristics of the population from which the sample was drawn. The quantitative component of this study was selected using a non-random sampling procedure. The quantitative design for this study was carried out using the purposive sampling technique, which was determined by the characteristics of the population and the study's goal. The researcher employed a homogeneous purposive sample in order to present a varied range of instances relevant to a specific phenomena or occurrence. As the name implies, the goal of this type of sample design is to provide the most amount of information feasible about the event or phenomena under investigation. Demographic data like Age, Gender, Occupation, Area of residency, Co morbid illness associated with Keratitis, Medications used, Trauma factors. The methods used for Specimen processing Wet mount, Potassium hydroxide Method, Grams staining, Culture and Sensitivity testing were performed.

RESULTS

400 were recruited for the study in accordance with the inclusion and exclusion criteria outlined in the protocol. According to the results of this study, keratitis patients 45%were mostly in the 41 – 50 year age range. In the participants, only5% of patients were beyond the age of 70, respectively. The gender distribution of the patients was analyzed. The study population consisted of 290 male patients (72.5%) and 110female patients (27.5%) respectively, showing a male preponderance. This table shows that, keratitis patients 72.5%were mostly from agricultural field. In the participants, only 5% of patients were unemployed.Table-4 It shows that 86.25%ofpatients from rural area randomly 13.75% of patients were from Urban area.

Table: 1. Prior Medication use of the Participants

Demographic Variables	Participants	
	No.	%
Medication		
Antimicrobials	118	29.5

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Antifungal	81	20.25
Corticosteroids	18	4.5
Antibiotics	43	10.75
No Medication	140	35
Total	400	100

Table 1 shows that 35% of the patients were not using any medication, while 4.5% were using Corticosteroids.

Table: 2. Risk factors of the participants

Demographic Variables	**Participants**	
	No.	**%**
Risk factors		
Trauma	322	80.5
Contact lens	16	04
Dry eye	35	8.75
No risk factors	27	6.75
Total	400	100

Table 2 shows that 80.5% of the patients associated risk factor were trauma, while 4.0 % were associated with the use of contact lens**.**

Table: 3. Causative agents identified among participants

Method	**Organism**	**Participants**	
		No.	**%**
culture	**Fungi**	322	80.5
culture	Bacteria	53	13.25
Wet mount	Parasites	13	03.25
PCR	Virus	12	03
Total		400	100

Table 3 shows that 80.5%of the were infected with fungi and only 3% clients were infected with virus. Its hows that 79%of the patients were positive in KOH Method, while 21 % were negative**.**

Table: 4. Association between Fungal culture and KOH method

	KOH+VE	**KOH-VE**		**chi-square**
				45.2204.P.< 0.00001
Culture+Ve	255	37	292	
Culture-ve	61	47	108	**significant at p < .05. Df-**
Total	316	84	400	

Table-4 It shows that KOH mount92.4% (292/316) was positive among participants , The chi-square value was 45.22 greater than table value (9.448) and is statistically significant atp<0.05.

Table: 5. Isolated Fungal species (n=316)

Fungus		**Participants**	
		No.	**%**
A	**Hyaline**		
1	A. fumigatus	78	24.68
2	A. flavus	58	18.35
3	A, Niger	42	12.29
4	F. solani	47	14.87
5	F. oxysporum	35	11.07
6	Penicillium species	36	11.39
B	**Dematiaceous**		
1	Bipolaris	8	2.53
2	Curvularia	8	2.53
3	Fonsaceapederosoi	4	1.26
		316	100

Table 5 shows that Majority 24.68% of the Isolated Fungal species are A. fumigatus. and the least isolated one was 1.26%Fonsaceapederosoi.Table-6Itshowsthat13.25%ofthepatients sample reveals the presence of bacteria , while 86.75 % were negative**.**

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Table 6 shows that 14% of the patients were positive in bacterial culture Method , while 86 % were negative**.**

Table: 6. Association between bacterial culture positivity and Gram staining method

	Gram stain +Ve	**Gram stain--Ve**		**chi-square**
				118.2032.
Culture+Ve	33	23	56	**P.< 0.00001**
Culture-ve	20	324	344	**significant at p < .05. Df-**
Total	53	347	400	

Table: 7. Isolated Bacterial organisms (n=53)

Fungus		**Participants**	
		No.	**%**
A	**Gram+Ve**		
1	S.aureus	30	56.60
B	**Gram-Ve**		
1	E.coli	4	7.55
2	K.pneumoniae	3	5.66
3	A.baumannii	1	1.88
4	P.aeruginosa	15	28.30
		53	100

It shows that Majority56.60% of the gram+Ve bacteria are S. aureus. and the least isolated one was 1.88% from gram -Ve.ieA. baumannii. It shows that wet mount 3.25% (13) was positive among participants , and none of them in culture positive.

Table: 8. Isolated parasitic organisms (n=13)

Virus		**Participants**	
		No.	**%**
A	**Acanthamoeba species**	13	100
		13	100

Table 8 shows that Acanthamoeba species was recognised from the direct smear of all (13)parasitic cases.

Table 9 Association between culture positivity and PCR method for Virus (n=400)

	PCR+Ve	**PCR--Ve**	
Culture+Ve	0	0	0
Culture-ve	12	388	400
Total	12	388	400

Table-9 shows that in PCR method 3.25% (12) was positive among participants , and none of them in culture positive.

Table: 10. Isolated Virus organisms

Virus		**Participants**	
		No.	**%**
A	**Herpes simplex virus**	10	83.33
B	**Varicella zoster virus**	02	16.67
		12	100

Table-10 shows that Majority 83.33% are Herpes simplex virus and the least isolated one was 16.67% from Varicella zoster virus.

Table: 11. Susceptibility of Natamycin and Ketoconazoleisolates

	Natamycin	**Ketoconazole**
Isolates	316	316
Susceptible	255	316
Resistant	61	

Table 11shows that 19.30% of the sample were resistance to Natamycinc But all samples were susceptible to ketoconazole.

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Table: 12. Quality control strains for Antimicrobial susceptibility

No	Antibiotic	P.aeruginosa 27853	MRSA 43300	E.coli 25922	MSSA 25923
1	GENTAMYCIN	21cm	20cm	23cm	25cm
2	CIPROFLOXACIN	28cm	24cm	30cm	29cm
3	MOXIFLOXACIN	NA	21cm	NA	25cm
4	ERYTHROMYCIN	NA	24cm	NA	29cm
5	DOXYCYCLINE	NA	26cm	NA	27cm
6	LINEZOLID	NA	27cm	NA	31cm
7	CLINDAMYCIN	NA	26cm	NA	31
8	COTRIMOXAZOLE	NA	25cm	26cm	31cm
9	CEFOXITIN	27cm	NA	NA	NA
10	PENICILLIN	NA	23cm	NA	38cm
11	AMPICILLIN	NA	NA	21cm	NA
12	AMIKACIN	24cm	NA	23cm	NA
13	CEFEPIME	28cm	NA	31cm	NA
14	AZTREONAM	27cm	NA	NA	NA
15	MEROPENEM	28cm	NA	31cm	NA
16	PIPERACILLIN- TAZOBACTAM	28cm	NA	NA	NA

Table: 13. Staphylococcus isolates-Antimicrobial susceptibilityn-30

No	Antibiotic	MSSA		MRSA	
		Resistant	Suspectable	Resistant	Suspectable
1	GENTAMYCIN		30	10	20
2	CIPROFLOXACIN	20	10	10	20
3	CHLORAMPHENICOL	30		5	25
4	ERYTHROMYCIN	20	10	20	10
5	DOXYCYCLINE	25	5	5	25
6	LINEZOLID		30		30
7	CLINDAMYCIN	20	10	10	20
8	COTRIMOXAZOLE	25	5	10	20
09	PENICILLIN	30		20	10

The table 13 reveals that S. aureus isolates MSSA and MRSA 50%.It also reveals that MRSA AND MSSA isolates linezolid100%and in MSSA , Chloramphenicol and penicillin were 100% resistant.

Table: 14. P. aeruginosa isolates-Antimicrobial susceptibilityn-15

No	Antibiotic		
		Resistant	Suspectable
1	Gentamycin	10	5
2	Amikacin	10	5
3	Cefepime	14	1
4	Aztreonam	-	15
5	Cefepime	14	-
6	Imipenem	14	1
7	Ciprofloxacin	10	5
8	Piperacillin-Tazobactam	10	5
09	Ceftazidime	5	10
10	Meropenem	-	15

Table: 15. E.coli isolates-Antimicrobial susceptibilityn-4

No	Antibiotic		
		Resistant	Suspectable
1	GENTAMYCIN		4
2	AMIKACIN		4
3	AMOXICILLIN- CLAVULANICACID		4
4	AMPICILLIN	4	4
5	CEFOTAXIME		4
6	IMIPENEM		

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7	CIPROFLOXACIN		4
8	COTRIMOXAZOLE	4	
09	CEFTAZIDIME		4
10	MEROPENEM		4

The table 15 revealed that E.coli isolates all antibiotics except Ampicillin and cotrimoxazole shows 100% resistance.

Table: 16. K. pneumoniae-Antimicrobial susceptibilityn-3

No	Antibiotic		
		Resistant	Suspectable
1	GENTAMYCIN	3	
2	AMIKACIN	3	
3	AMOXICILLIN- CLAVULANICACID	3	
4	CEFOTAXIME	3	
5	IMIPENEM		3
6	CIPROFLOXACIN		3
7	COTRIMOXAZOLE	3	
8	CEFTAZIDIME	3	
9	MEROPENEM		3

The table 1 6 reveals that K. pneumoniae susceptible to three medicines, they are Imipenem, Ciprofloxacine and meropenem and all other medicines shows 100% resistance.

Table: 17. A.baumannii-Antimicrobial susceptibilityn-1

No	Antibiotic		
		Resistant	Suspectable
1	GENTAMYCIN		1
2	AMIKACIN		1
3	AMOXICILLIN- CLAVULANICACID		1
4	CEFOTAXIME		1
5	IMIPENEM		1
6	CIPROFLOXACIN		1
7	COTRIMOXAZOLE	1	
8	CEFTAZIDIME		1
9	MEROPENEM		1
	AMPICILLIN	1	

The table 17 reveals that K. pneumoniae susceptible to all medicines, except cotrimoxazole and ampicillin shows 100% resistance.

Table: 18. Acanthamoeba species-Antiparasitic susceptibilityn-13

No	Antiparasitic		
		Resistant	Suspectable
1	Chlorhexidine	-	13
2	Pentamidineis ethionate	-	13
3	Diminazene aceturate	3	10

The table 18 reveals that Acanthamoeba species susceptible to Chlorhexidine and Pentamidine is ethionate and Diminazene aceturate shows some resistance.

Table: 19. Herpes simplex virus-Antiviral susceptibilityn-10

No	Antiviral		
		Resistant	Suspectable
1	Idoxudine	3	07
2	Trifluridine	1	09
3	Acyclovir	-	10

The table 19 reveals that Herpes simplex virus susceptible Acyclovir and Idoxine and Trifluridine shows some resistance.

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Table: 20. Varicella zoster virus-Antiviral susceptibilityn-2

No	Antiviral		
		Resistant	Suspectable
1	famciclovir		02
2	penciclovir		02
3	Acyclovir	01	01

The table 20 reveals that Varicella zoster virus susceptible to famciclovir and penciclovir and Trifluridine shows 50%resistance.

DISCUSSION

Regarding injudicious use of medication,35% of the patients were not using any medication, while20.5 antifungal,29.5% antimicrobial 4.5%and 10.75% uses antibiotics were using Corticosteroids. A similar outcome was found in the investigation of (Noopur Gupta .2017)[7]Results show that out of the 2160 people surveyed, 396 (18.2%) admitted to self-medicating with ophthalmic drugs to treat symptoms including watering (37.1%), redness (27.7%), itching (19.2%), and infection (19.2%). (13.6 percentage point). When eye drops were made available without a prescription, 26.4 percent of subjects were found to be self- medicating when they were physically checked.151 (26.5%), 120 (21.1%), and 75 (13.2%) subjects reported using steroids, expired/unlabeled, and indigenous eye drops ,respectively. Some 529 participants also used home treatments such as "kajal"(61.4percent),honey(31.4percent),ghee(11.7per cent) and rose water(5.7percent)(9.1percent).The study investigated risk factors and characteristics of keratitis patients, revealing that 80.5% were associated with trauma and 4% with contact lens use, consistent with prior findings by Stefan and Nenciu.[8] Analysis of 68 cases showed that corneal scraping was performed in 96% of cases, with 86% positive results. Gram-positive bacteria were involved in 76% of cases, with Staphylococcus epidermidis being the most common isolate. Gram-negative bacteria were linked to severe anterior chamber inflammation. Antimicrobial treatment was modified in 13.2% of cases, with 95.5% of ulcers cured, though only 75% of patients had improved visual acuity. Fungal infections predominated, with Aspergillus flavus being the most common fungus. Voriconazole was highly effective against mold keratitis, while fluconazole showed resistance. The study by Green et al.[9] reviewed 257 microbial keratitis cases, identifying contact lens wear, ocular surface illness, and trauma as risk factors. Pseudomonas aeruginosa was common among contact lens wearers, and Fusarium and P. aeruginosa infections were associated with severe keratitis. The KOH method detected fungal infections in 79% of patients, and Aspergillus species were the most common fungi. Abouzeid's et al [10]study found fungal growth in 46% of cases, with Aspergillus spp. predominating. Voriconazole was effective, while fluconazole was not recommended for empirical therapy. Bacterial staining showed 13.25% of samples had bacteria, with S. aureus being the most common Gram-positive isolate. Antifungal susceptibility testing revealed high sensitivity to ketoconazole. Peterson's[11] study highlighted the resistance profiles of MRSA and MSSA keratitis isolates, with USA300 and USA100 being the most common strains. Acanthamoeba species were sensitive to chlorhexidine and propamidine. Bacon et al. noted that acyclovir resistance in herpes simplex virus remained low despite extensive use, but higher in immunocompromised individuals.[12] This comprehensive analysis underscores the importance of accurate diagnosis and appropriate treatment to manage keratitis effectively.

CONCLUSION

Fungal corneal ulcers were found to be more common than bacterial corneal ulcers in this study. Because of our country's tropical environment, which is both warm and humid, this is understandable. Simple bedside diagnostics, such as direct Gram staining or KOH mount and PCR test can provide valuable information on the type of the infectious agent and can assist doctors in selecting the most appropriate antibiotic treatment.

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