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Bacteriological Profile of Keratitis and It's Antibiotic Susceptibility Pattern

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ABSTRACT

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INTRODUCTION

Keratitis is an inflammation of the cornea caused by infectious organisms or non-infectious agents or stimuli. Infectious keratitis may be caused by bacteria, viruses, fungi and parasites. Keratitis often causes corneal scarring and opacification.[1]

Corneal infections are the second most common cause of monocular blindness after unoperated cataract in developing countries. Surveys in Asia and Africa have confirmed these findings. [2-4] Corneal opacities, which are largely caused by infectious keratitis, are the fourth leading cause of blindness globally and are responsible for 10% of avoidable visual impairment in the world's least developed countries.[5,6] Approximately 2 million people develop a corneal ulcer every year in India alone.[7,8]

The majority of bacteria cultured from the infection of the cornea are of same species that normally present in conjunctival sac, on the lids or periocular skin.[9] The most common bacterial species isolated are staphylococcus aureus (25.13%) followed by streptococcus pneumonia (21.78%) and coagulase-negative staphylococcus aureus.[10]

Considering importance of corneal ulceration as an important cause of visual loss, many studies have reported prevalence of microbial pathogens and identified the risk factors predisposing a population to corneal infection in south India[11]. The present study is undertaken to determine the bacteriological profile of keratitis and their antibiotic susceptibility pattern.

OBJECTIVES

1. To identify the pathogens responsible for bacterial keratitis.
2. To determine their antibiotic susceptibility pattern.

COLLECTION OF SAMPLES

Corneal scrapings are collected under strict aseptic precautions by an ophthalmologist using a sterile number 5. Bard Parker blade after instillation of a local anaesthetic like 2% lignocaine hydrochloride.Materials obtained from scraping the leading edge and base of each ulcer was smeared onto two slides for direct microscopic examination and inoculated onto culture medias[12]

SOURCE OF DATA:

The present study was carried out in Dept. of Microbiology, VIMS hospital, Ballari, from January 2017 to December 2017 and 100 samples were collected.

The material for the present study was obtained from patients in the Opthalmology Department, who

presented to the OPD with signs & symptoms of keratitis.

Inclusion criteria:

Patients presented with suspected infectious bacterial keratitis to the OPD.

Exclusion criteria

Patients suspected of having or with a positive cultures for anaerobic bacteria, fungal, viral or parasitic infections.

METHODS OF COLLECTION OF DATA:

All the patients underwent slit lamp bio-microscopic examination by an ophthalmologist. After a detailed ocular examination, using standard technique, corneal scrapings was taken under aseptic conditions by an ophthalmologist using a sterile bard-parker blade (No. 5). The procedure was performed under the microscope after instillation of 4% lignocaine without preservative.

The material scraped was inoculated into the surface of solid media such as blood agar, chocolate agar. The material obtained by scraping was also spread onto labeled slides in a thin even manner to prepare 10% potassium hydroxide wet mount.I have excluded positive KOH samples.

Bacterial cultures were considered positive only if growth of the same organism is demonstrated on both media or if there is semi confluent growth at the site of inoculation on one media with identification of morphological characteristics of similar organism in Gram Stain. The specific identification of bacterial pathogens were based on microscopic morphology, staining characteristics and biochemical properties using standard laboratory criteria.

The bacterial strains were tested for anti-microbial susceptibility by Kirby Bauer Diffusion method according to CLSI guidelines.

RESULTS

One hundred patients clinically diagnosed cases of corneal infections attending Opthalmology outpatient department and those admitted in Opthalmology wards were studied in the Department of Microbiology , Vijaynagar institute of Medical Sciences,Ballari. Observations made from the study are as follows.

Age of the subjects in the study group varies from 12 years to 74 years. Cases presented to the OPD with corneal infection symptoms were common in the age group of 51-60 years.

Age – Sex wise distribution of patients

Age group(yrs)	Sex		Total
	Male	Female	
< 20	2	1	3
21-30	3	4	7
31-40	14	12	26
41-50	16	11	27
51-60	14	14	28

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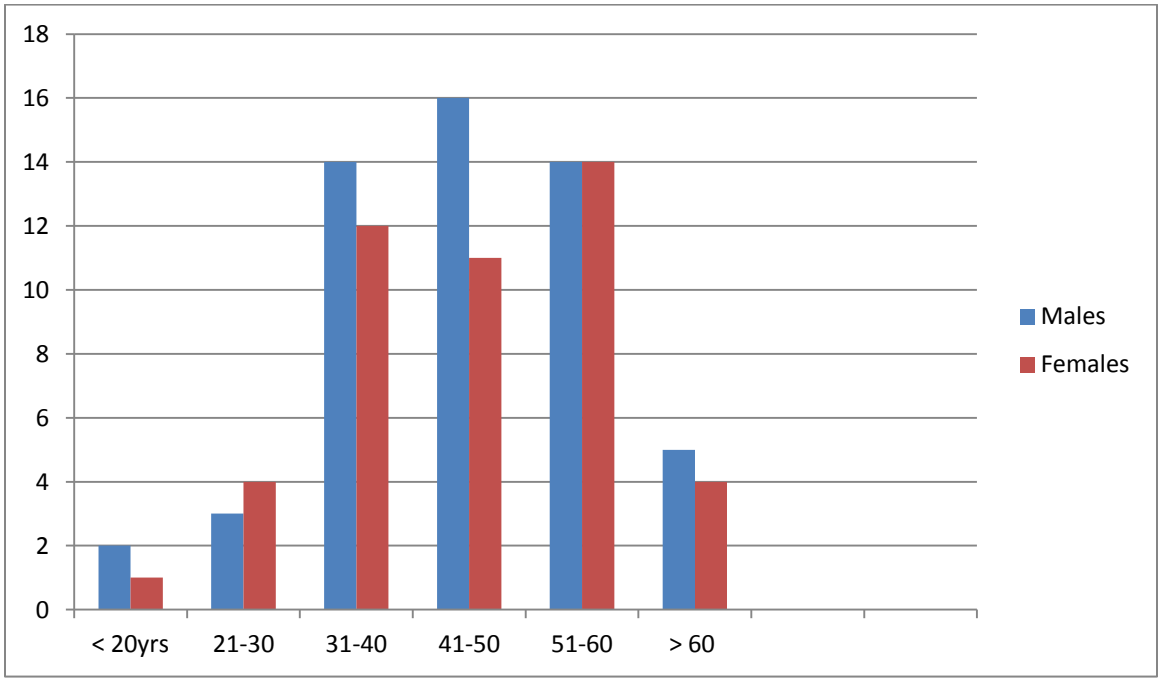
>60	5	4	9
Total	54	46	100

Age – Sex wise distribution of patients



Distribution of patients based on area

Locality	Frequency	Percentage
Rural	77	77%
Urban	23	23%
Total	100	100%

Out of 100 cases, 77 cases were from rural areas and 23 cases were from urban areas. Above table shows corneal infections were common in rural areas compared to urban areas.

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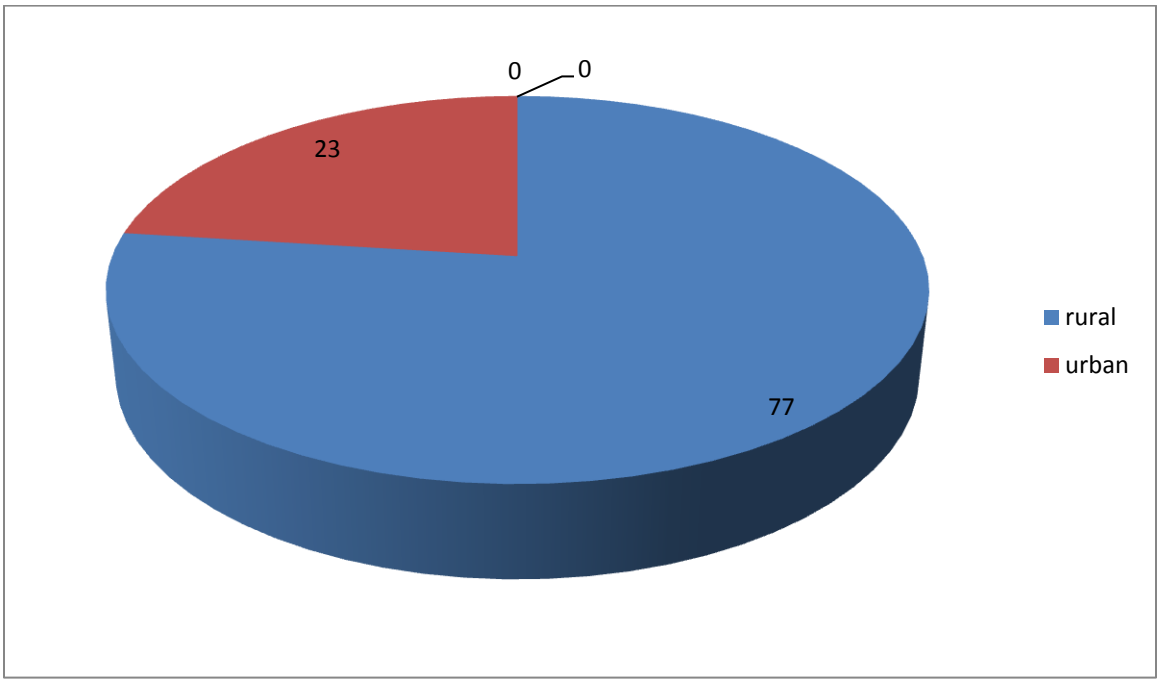
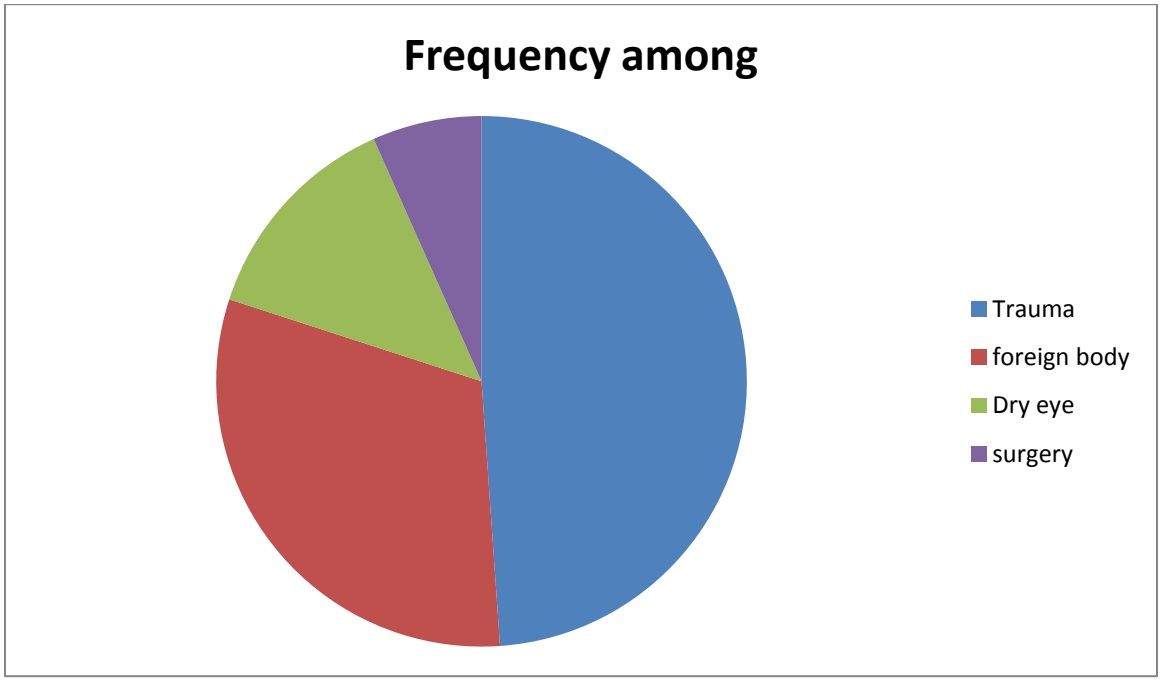
Graph 3 : Distribution of patients based on area



Graph 4 : Risk factors among patients in the study group



Out of 100 patients presenting with complains of ocular infections , 22 patients had history of trauma, 14 of them gave the history of foreign body, 6 cases had dry eye and 3 patients with a surgical history.

Distribution of patients based on occupation

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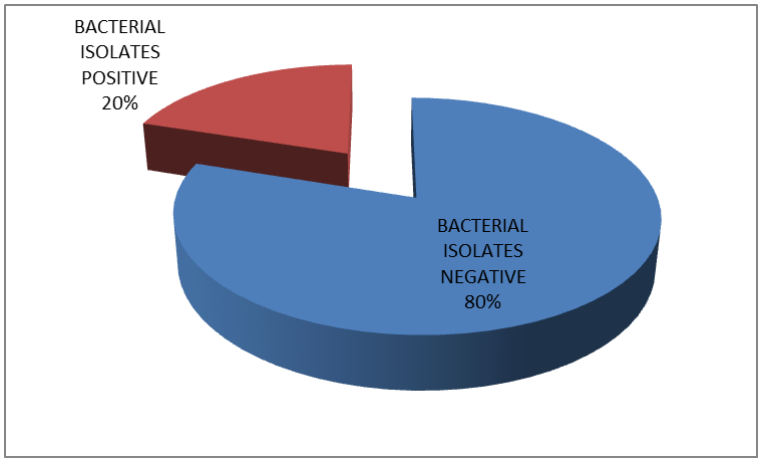
Occupation	Frequency	%
BUSINESS	6	6.0
FARMER	39	39.0
H/W	6	6.0
LABOUR	27	27.0
OTHER	6	6.0
STUDENT	5	5.0
TAILOR	3	3.0
VENDOR	8	8.0
Total	100	100.0

The above table shows that corneal infections were common in farmers (39%) and manual labours (27%).

Graph 5: Bacterial culture results



The above table shows that out of 100 samples, 20 (20%) were culture positives and 80 (80%) were culture negative.

Relation between age and culture positive results

AGE GROUP (years)	CULTURE POSITIVE
< 21	0

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21-30	1(5%)
31-40	7(35%)
41-50	5(25%)
51-60	6(30%)
> 60	1(5%)

Chi- square test = 12.01 p = 0.7

Culture positivity was more common in 31-40 years of age group and this can be attributed to increased outdoor activities in this age group.

Relation between sex and culture positive results

Sex	Culture positive	Culture negative	Total	Incidence
Male	14	42	56	25%
Female	6	38	44	13.63%
Total	20	80	100	

Chi-square test = 1.9886 p = 0.158

The above table shows bacterial keratitis has more male preponderance 14 cases than female 6 cases and can be attributed to more outdoor activities in males in sub-continents which is statistically not significant.

Relation between area and culture results

Area	Culture positive	Culture negative	Total	Incidence
Rural	15 (75%)	62	77	19%

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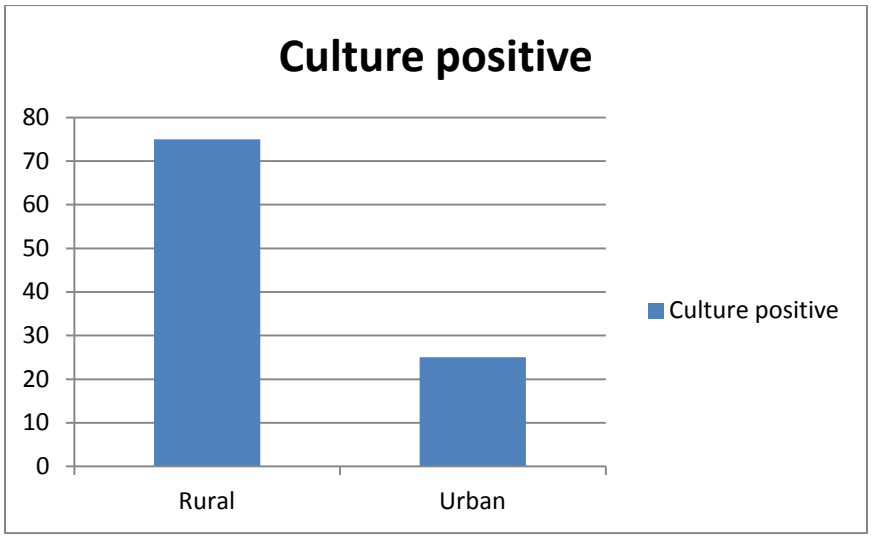
Urban	5 (25%)	18	23	23%
Total	20 (100%)	80	100	

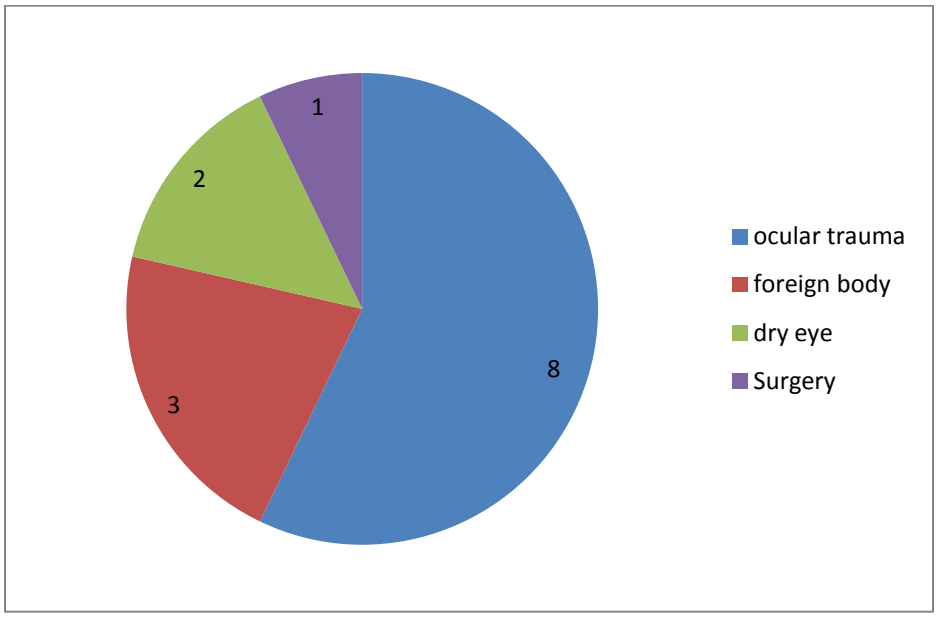
Chi-square test = 0.05 p = 0.81

The above table shows high culture positivity(75%) in rural cases as compared to urban cases (25%) whereas incidence is higher in urban than rural which is statistically not significant.

Relation between area and culture results



Risk factors among culture positive patients



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Ocular trauma (40%) and foreign body (15%) were the common predisposing factors for bacterial keratitis. Other risk factors were dry eye and surgeries.

Occupation wise culture positive bacterial keratitis

OCCUPATION	FREQUENCY	PERCENTAGE
Farmer	12	60%
Labour	5	25%
Housewife	1	5%
Student	1	5%
Vendor	1	5%
Total	20	100%

Bacterial keratitis was more common in agriculture workers (60%), followed by manual labours (25%).

Pattern of bacterial isolates

Bacterial isolates	Frequency	percentage%
S.AUREUS	8	40.0
CONS	7	35.0
S.PYOGENS	3	15.0
Pseudomonas	1	5.0
Acinetobacter	1	5.0
Total	20	100.0

A total of 5 bacterial species were isolated from 20 culture positive cases. The above table shows that the most common organism isolated was *S.aureus (40%) followed by CONS (35%), S.pyogenes (15%), Pseudomonas (5%) and Acinetobacter (* 5%).

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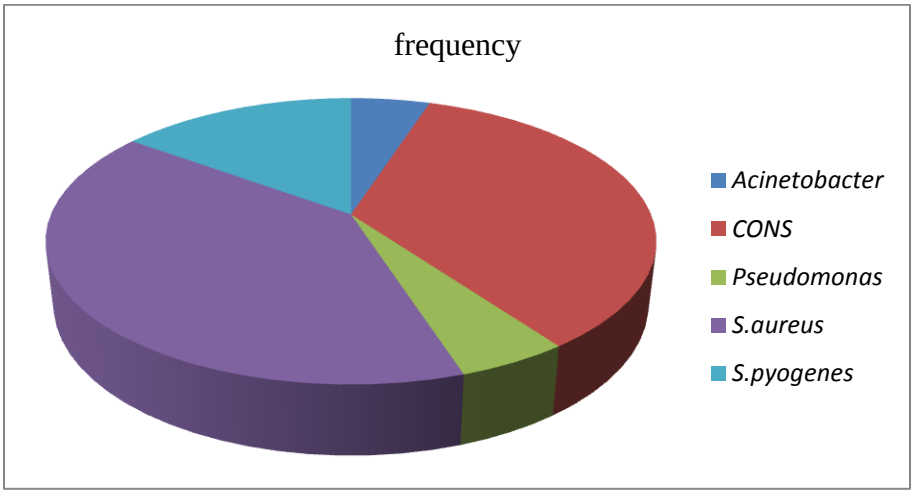
Pattern of bacterial isolates



ANTIBIOTICS	S. aureus (n = 8) sensitivity	CONS (n = 7)	S.pyogenes (n = 3)	Pseudomonas (n = 1)	Acinetobacter (n = 1)
Amikacin	6 (80%)	6 (85%)	3 (100%)	1 (100%)	R
Ciprofloxacin	2 (25%)	5 (71%)	3 (100%)	1(100%)	R
Cotrimoxazole	1 (12.5%)	3 (43%)	1 (33.3%)	1(100%)	1(100%)
Clindamycin	1 (12.5%)	4 (60%)	3(100%)	1(100%)	1(100%)
Erythromycin	2 (25%)	3 (42%)	3(100%)	1(100%)	R
Vancomycin	8 (100%)	7 (100%)	3(100%)	0	0
Cefotaxime	6 (75%)	3 (42%)	1(100%)	1(100%)	1(100%)
Amoxiclavulate	8 (100%)	7 (100%)	1(100%)	R	R
Ampicillin	7 (87%)	6 (85%)	3(100%)	R	R

ANTIMICROBIAL SENSITIVITY PATTERNS OF ISOLATED BACTERIAS

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DISCUSSION

Bacterial keratitis is an ophthalmic emergency that needs immediate institution of treatment. In absence of laboratory diagnosis the initial therapy is usually broad spectrum intensive treatment.

Specific therapy should be based on laboratory data which identifies the causative agent and provides antibacterial susceptibility results.

In the present study the bacterial culture were obtained from 23% of patients with corneal infections. Similar observations were made in the following studies.

AK Leck. Et al reported bacterial etiology in 29% of the patients with infectious keratitis during 1999-2001 out of 800 cases from South India [12].

The study done by S. Krishna et al in the year 2015 on 300 patients revealed bacterial etiology in 20.7% patients[13]. Jadhav S.V et al studied 271 patients with corneal ulceration at a tertiary care hospital in western India during the period of 2007-10 and found their incidence to be 25.9%[1].

MJ Bharathi et. al performed retrospective review of medical records of all culture-positive bacterial keratitis which were seen over a 3 years period, from September 1999 through August 2002 at a tertiary care referral centre in South India. In their study the incidence of bacterial etiology was 32.7 %[9]

Age/Sex wise distribution of bacterial keratitis In the present study males (54%) were affected more than females (46%). The most common affected age group was 31-40 years in both sexes.

Males were more commonly affected and the majority of the patients were in the age group of 31-40 years, in a study by Ragini Tilak et al. at Banaras University, Varanasi[14] .Similarly males (67.6%) were commonly affected than females(32.4%) and the most commonly affected age group was 41-50 years (25.5%) in a study done by Reema Nath et al from Assam[15].

Area wise distribution

In the present study, cases of corneal infections were more common rural areas (77%) than patients from urban area (23%).In our study incidence of keratitis was more in urban population (23%) as compared to rural (19%) but the study was not significant. Our study results differed from following studies,

A population-based cross-sectional study of 5150 persons 40 years or older in a randomly chosen rural population of 3 districts of southern India by Praveen K, et al. concluded that bacterial keratitis was more prevalent in rural areas than urban which is in accordance with our present study [16].

MJ Bharati et al. in a tertiary referral centre in south India isolated 1109 bacterial pathogens and concluded that the cases were from more of rural population (54.6%) in comparison to urban population[9]. M Srinivasan et al. evaluated 434 patients in a 3 month period with high risk factor of corneal injuries causing ulceration were more from rural areas as compared to urban populationThis variation in our study may be attributed to low sample size and location of the study hospital in urban area where in patients from rural areas may be have taken treatment from primary health centres around them.

Predisposing factors

Most of the studies done on bacterial keratitis have listed trauma , contact lens wear and dry eye as important risk factor occurring in 70-75% of the patients. Other risk factors include foreign body, surgeries, ocular surface pathologies, corneal exposure in coma and general anaesthesia patients. However there is a significant difference in risk factors among rural and

urban population.

In the present study, trauma(40%) and foreign body(15%) were the common predisposing factors for bacterial keratitis. Other factors were dry eye and surgery.

Reena Gupta et al. evaluated 40 culture positive cases from Jammu and found that ocular trauma was most important risk factor (80%) causing corneal ulcers[17].

Occupation wise distribution

In the present study, bacterial keratitis was commonly observed in farmers(60%) followed by manual labour(25%). Our study holds good in accordance with the following various studies.

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A majority of the patients with bacterial keratitis were farmers (64.7%) in a study done by M.Jayahar Bharathi et al from south India[9].

Reena Gupta et al. after evaluating 41 cases from Jammu concluded that males in rural agricultural population in economically productive age group are most vulnerable to infective keratitis [17] .

Various bacterial agents isolated

In the present study the most common organisms isolated were S.aurues (40 %) followed by CONS (35%), S.pyogenes (15%) ,Acinetobacter (5%) and Pseudomonas (5%).

A similar study was conducted by Arti Tewari on 150 patients in a year in Ahmedabad and found that S.aureus (32%) was the most commonly isolated organism followed by CONS(25%)[18].

S.Marisini et al. with the objective of isolating the most common bacteria causing bacterial keratitis in New Zealand conducted a study over 24 months from Jan 2013 to Dec 2014 isolated 126 culture positive cases and found that S.aureus was the most common agent with 38%

incidence followed by pseudomonas[19].

In a study conducted by Amrutha kumara et al. on consecutive 100 patients found

Patricia Chirinos-Saldaña et al. conducted a retrospective review of clinical records of patients under 16 years of age with history of microbial keratitis seen at a tertiary referral center and found Gram-positive bacteria were isolated in 78.5%; Staphylococcus epidermidis (28.6%) was the most common microorganism[20].

ANTIBIOTIC SENSITIVITY OF ISOLATED ORGANISMS

In the present study it is evident that the most effective antibiotic for the treatment of Gram-positive cocci infections (caused mostly by S.aureus,CONS and S.pyogenes)are Vancomycin and

Amoxiclavulanate effective against 100 % bacteria.

The commonest bacteria isolated in this study, Staphylococcus aureus showed 100 % sensitivity to Vancomycin and Amoxiclavulanate, 87% to Ampicillin, 80% to Amikacin, 75 % to cefotaxime, 25% to Erythromycin and Ciprofloxacin. In a study by Khosravi A D et al[21] and Anisha Et al[22]., similar to our findings Staphylococci showed high sensitivity to Vancomycin.

The second commonest isolate CONS showed 100 % sensitivity to Vancomycin and Amoxiclavulate, around 75% sensitivity to Ciprofloxacin and poor sensitivity to Erythromycin ,

Cefotaxime and Cotrimoxazole.

H.O.Orlans in a 10 year review from 1999-2009 in Oxford, UK also showed the similar results to our study in which CONS was 100 % sensitive to vancomycin and around 85% sensitive to

Ciprofloxacin [23].

In the present study 5% each of Pseudomonas and Acinetobacter were isolated and were 100 % susceptible to Ciprofloxacin, Cefotaxime, Amikacin and showed resistance to penicillin containing antibiotics like Ampicillin and Amoxiclavulanate.

CONCLUSION

The study on bacterial keratitis showed Staphylococcus aureus as the most common causative agent followed by Coagulase negative Staphylococcus. Other bacterial isolates were S.pyogenes, Acinetobacter and pseudomonas.

Bacterial keratitis was commonly seen in young male patients engaged in agricultural related work and outdoor work like manual labours. The common predisposing factors were trauma and foreign bodies.

Isolated organisms(Gram positive cocci) showed high sensitivity to Vancomycin and variable sensitivity to other penicillin group of drugs and fluoroquinolones. On the other hand Gram negative bacilli showed resistance to cephalosporins, fluoroquinoles which are the common antibiotics used in general settings.

Bacterial keratitis continues to be an important cause of ocular morbidity, mostly in the persons living in rural areas, involved in outdoor agricultural activity. Young male adults affected in these circumstances are often from low socioeconomic status and the sole bread earners of their families. Hence blindness will be of great economic burden with grave consequences. Therefore ,spreading awareness about the early diagnosis and prompt treatment with susceptible antibiotics may limit mortality.

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SUMMARY

One hundred patients with corneal ulcers attending Ophthalmology outpatient department and admitted in Ophthalmology wards were studied in the Department of Microbiology, Vijayanagar Institute of Medical Sciences, Bellary, to know the common bacterial organisms causing keratitis, predisposing factors for keratitis. In the present study the antibiotic susceptibility pattern was also analysed.

1. Age of the patients ranged from 12 years to 74 years. Maximum incidence of culture positive cases were found during 30-50 years (60%)
2. Maximum number of the patients were males (54%) than the females (46%)
3. Patients from rural area (77%) were commonly affected than urban area (23%)
4. The common predisposing factors were trauma, foreign bodies and agricultural related occupation
5. Total number of sample studied were 100, of which 23% were culture positive.
6. A total of 5 bacterial species were isolated from 20 culture positive cases. The most common organism isolated was *Staphylococcus aureus* (40%) followed by *CONS* (35.0%).

Gram positive cocci were highly sensitive to Vancomycin and Amoxiclavulanate where as gram negative bacilli were resistant to commonly used penicillin group of drugs.

REFERENCES

1. Jadhav SV Et. Al. Prevalence of fungal keratitis from tertiary care hospital from western part of India. Int J Microbial res. 2012;4(4):211-214
2. Upadhyay MP, Karmacharya PC, Koirala S, Tuladhar NR, Bryan LE, Smolin G, et al.Epidemiologic charactersiticcs, predisposing factors and aetiologic diagnosis of corneal ulceration in Nepal. Am J Opthalmol 1991; 111:92-99
3. Gonzales CA, Srinivasan M, Whitcher JP, Smolin G. Incidence of corneal ulceration in Madurai District,south India.Opthalmol Epidemiol 1996;3:159-66
4. Whitcher JP, Srinivasan M, Corneal Blindness:a global perspective.Bull World Health Organ 2001;79:214-21
5. Pascolini D, Mariotti S P.Global estimates of visual impairment:2010 Br ophthal 2012;96:614-618 Lancet. 1991;338:650-653.
6. World health organisation. Causes of blindness and visual impairment. Available at http://www.who.int/blindness/causes/en.Acce sses December 7, 2016.
7. Gupta N, Tandon R, Gupta SK, et al. Burden blindness in India. Indian J community Med.2013;38:198-206.
8. Dart JK, Stapleton F, Minassian D. Contact lenses and other risk factors in microbial keratitis. Lancet. 1991;338:650-653.
9. M J Bharathi, R Ramkrishna, S Vasu, Meenakshi, R Palaniappan. Aetiological diagnosis of microbial keratitis in south India-A study of 1618 cases. IJMM 2002;20:19-24
10. S.Ramesh, R.Ramkrishnan, M.Jayahar, Bharathi M, S. Vishwanathan. Prevalence of bacterial pathogens causing ocular infections in south India . Indian journal of Pathology and Microbiology. 2012; 53(2)281-286
11. S.Smitha, P Lalitha, VN Prajna, M Srinivasan. Susceptibility trends of pseudomonas species from corneal ulcers. IJMM 2005;23(3):168-171
12. Leck AK, Thomas PA, Hagan M, Kaliamurthy J, Ackuaku E, John M et al. Aetiology of suppurative corneal ulcers in Ghana and south India and epidemiology of fungal keratitis. Br J Opthalmol 2002;86:1211-15
13. Waxman E, Chechelnitsky M, Mannis MJ, Schwab IR. Single culture media in infectious keratitis. Cornea 1999;18:257-61.
14. Tilak R, Singh A, Maurya O P S, Chandra A, Tilak V, Gulati A K. J Infect Dev Ctries.2010;4(3):171-174

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15. Nath R, Baruah S, Saikia L, Devi B, Borthakur A K, Mahanta J. Mycotic corneal ulcers in upper Assam. Indian J Ophthalmol.2011; 59(5):367-371

16. Houang E, Larn D, Fan D, Seal D. Microbial keratitis in Hong Kong: relationship to climate,environment and contact-lens disinfection. *Trans Roy Soc Trop Med Hyg* 2001;**95**:361-367.

17. Schaefer F, Bruttin O, Zografos L, Guex-Crosier Y.Bacterial keratitis: a prospective clinical and microbiological study. *Br J Ophthalmol* 2001;**85**:842-847.

18. McDonnellPJ, Nobe J, Gauderman WJ, Lee P, Aiello A, Trousdale M. Community care of corneal ulcers. Am J Ophthalmol 1997;115:462-5

19. Courtright P, Lewallen S, Kanjaloti S. Changing patterns of corneal disease and associated vision loss at a rural African hospital following a training programme for traditional healers.Br J Ophthalmol 1996;80:694–7.

20. Marangon FB, Miller D, Alfonso EC. Impact of prior therapy on the recovery and frequency of corneal pathogens. Cornea 2004;23:158-64

21. A.D Khosravi, F.Ahmadi, S.Salmanzadeh, A.Dashtbozorg. Study of bacteria isolated from Orthopedic implant infections and their antimicrobial susceptibility pattern. Research journal of Microbiology.2009;4:158-163

22. Anisha Fernandes, Meena Dias. The Microbiological profiles of infected Prosthetic implants with an emphasis on the organisms which form biofilms. Journal of Clinical and Diagnostic research. 2013;7(2):219-223

23. HO Orlans1, SJ Hornby1 and ICJW Bowler. In vitro antibiotic susceptibility patterns of bacterial keratitis isolates in Oxford, UK: a 10-year review. Eye (2011); 25: 489–493

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