

Original research article

## Microbiological Profile and Antibiotic Sensitivity Pattern of Ocular Infections in A Tertiary Care Hospital

Dr. Rakhee Agarwal

Assistant Professor, Department of Microbiology, Prathima Institute of Medical Sciences, Nagunur, Karimnagar.

Corresponding Author: Dr. Rakhee Agarwal

E-mail: rakhee\_micro21@gmail.com

### Abstract

**Background:** The eye may become infected from the outside or as a result of bloodstream-borne germs invading the eye. In this work, particular bacterial and fungal pathogens that cause eye infections were isolated, identified, and their antibiotic susceptibilities were assessed. This research was done to find the bacterial and fungal pathogens that cause eye infections, isolate and identify them, and find out how susceptible they are to antibiotics in vitro.

**Methods:** All of the participants in the current study had slit lamp bio-microscopy examinations, and an ophthalmologist made a clinical diagnosis based on accepted practises. The various ocular tissues were collected for specimens for smear and culture after thorough ocular examinations utilizing conventional methods. The samples were examined under a microscope directly using techniques like Gram staining, and biochemical reactions performed were catalase test, coagulase test Motility test, Indole test, Citrate utilisation test, Urea Hydrolysis test, and sugar fermentation tests.

**Results:** In vitreous and corneal specimens, growth was observed in n=20 (32.25%) of the n=26 (41.93%) instances that were found to be positive by direct microscopy (KOH mount with Calcofluor White staining), while no growth was found in n=6 cases. Out of n=36 (58.06%) cases that were negative on direct microscopy, growth was seen in n=2 (5.55%) cases, whereas no growth was seen on culture in the remaining n=34(94.44%) cases. Direct microscopy had a sensitivity of 80.12% and a specificity of 79.61%. Out of total n=20 fungal isolates found n=15 cases were from corneal scrapings which yielded *Aspergillus sp.* In n=6, *Penicillium sp.* n=5 *Candida albicans* n=2 and *Fusarium sp.* n=2 and the one mixed growth of candida along with Coagulase negative staphylococcus.

**Conclusion:** Emerging drug resistance is a matter of serious concern and hence all ophthalmological samples must be analysed as thoroughly as possible for antimicrobial resistance due to the increase of antibiotic resistance. Ophthalmologists must adhere to the etiologic approach to diagnosis and consider risk reduction in order to lessen the burden of ocular infections.

**Keywords:** Ocular Infections, Antibiotic Resistance, Corneal Scrapings, Fungal Kertomycosis

### Introduction

One of the infections that is most frequently encountered is ocular infection. Normally, the eye is protected against infections by a number of natural defence mechanisms. These include the corneal surface epithelium, the blink reflex, and the bioactive lysozyme, IgA, and IgG

components of the tear film. <sup>[1]</sup> When these defences are breached by exogenous or endogenous conditions that enable microbial invasion of the eye, infection occurs. Exogenous infection can develop as a result of intraocular surgery or penetrating eye damage. As a result of the haematogenous transmission of infection from other bodily areas, infection is acquired endogenously. The conjunctiva, cornea, and eyelids are the parts of the eye that are most frequently affected. <sup>[2]</sup> Visual impairments caused by even a mild illness elsewhere in the body might can be devastating to the eye. Any area of the eye is susceptible to bacterial, fungal, parasitic, or viral infections. <sup>[3, 4]</sup> Due to the extensive use of topical, systemic immunosuppressive drugs and the rising number of HIV patients, many opportunistic agents are frequently seen in ocular infections. <sup>[5]</sup> Around the world, bacteria are the main cause of ocular illnesses. Contact lenses, trauma, surgery, dry eye, chronic nasolacrimal duct obstruction, and prior ocular infections are just a few of the variables that might cause an infection, which can be mono- or poly-microbial. <sup>[6]</sup> The edges of the eyelids are home to several bacteria that spread diseases. Although they typically only affect a small area, these infections can occasionally spread to nearby tissues including the conjunctiva and cornea. The main culprit behind infections of the eyelids is bacteria. *Staphylococcus aureus*, *Streptococcus species*, and *Pneumococcus* etc several kinds of Streptococcus, Pneumococcus, and other organisms are frequently involved. Dacryocystitis is inflammation of the lacrimal sac and occurs due to blockage of secretion of the tears. This causes accumulation of secretions and tears within the sac and causes infection. The organisms causing these infections are mainly *Staphylococcus aureus* and *Streptococcus species* which usually arise from the conjunctival sac as they are seen as commensals. This is of particular importance since if left untreated it may lead to spread of infections to other parts of the eye. <sup>[7]</sup> Corneal ulcerations brought on by bacterial keratitis results in corneal opacity and severe vision impairment. <sup>[8]</sup> Pyogenic organisms including *Staphylococcus aureus*, *Pneumococcus*, *Pseudomonas aeruginosa*, *Escherichia coli*, etc. are mostly to blame for the exogenous nature of the infection. Due to a flaw in the corneal epithelium, fungi can enter the cornea and cause tissue necrosis, which in turn causes ulceration and corneal opacity. Due to its protracted course and prevalence among rural agricultural labourers, mycotic keratitis has a poor prognosis and is a significant cause of blindness. <sup>[9]</sup> With this background this study, the bacterial and fungal pathogens that cause eye infections were isolated, identified, and their invitro susceptibilities to common antibiotics were assessed.

### Material and methods

This cross-sectional study was conducted in the Department of Microbiology with the support of the Department of ENT, Prathima Institute of Medical Sciences, Nagunur, Karimnagar. Institutional Ethical approval was obtained for the study. Written consent was obtained from all the participants of the study. All of the patients in the current study had slit lamp biomicroscopy examinations, and an ophthalmologist made a clinical diagnosis based on accepted practises. The various ocular tissues were collected for specimens for smear and culture after thorough ocular examinations utilizing conventional methods. The samples were examined under a microscope directly using techniques like Gram staining, and biochemical reactions performed were catalase test, coagulase test Motility test, Indole test, Citrate utilisation test, Urea Hydrolysis test, and sugar fermentation tests. A 10% KOH wet mount, Calcofluor white staining, for identification of fungi. After that, the samples were placed on Blood, MacConkey, and Chocolate agar plates, incubated aerobically for 18–24 hours, and then inspected the next day. Additionally, duplicate samples of the specimens were inoculated onto Sabouraud's dextrose agar, with one batch incubated at room temperature and the other at 37 °C. SDA slopes were evaluated twice weekly for the following three weeks after being examined daily for the first week. The usual microbiological techniques were used to identify isolated bacteria and

fungus. The recent CLSI recommendations (2018) were followed for evaluating the bacterial isolates for antimicrobial susceptibility. <sup>[10]</sup>

## Results

A total of n=110 samples were collected comprising of n=67(60.91%) conjunctival swabs, n=34(30.90%) corneal swabs, n=24(21.81%) lacrimal pus samples and n=28(25.45%) vitreous samples. Out of the samples n=61 showed growth and n=55(90.16%) were bacterial growth and n=6(9.8%) were fungal isolates. Out of n=110 samples collected n=70 were males and n=40 were females. The mean age of the cases in the study was  $44.58 \pm 11.25$  years the details of the distribution of the cases age wise is given in table 1.

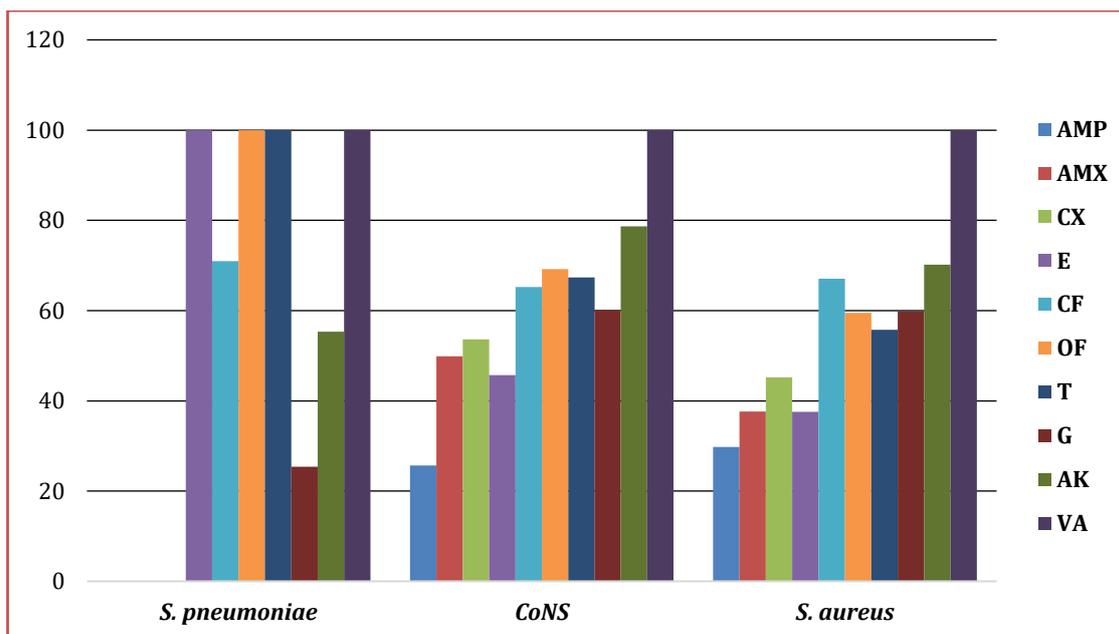
**Table 1: Demographic profile of the cases included in the study**

<i>Age in years</i>	<i>Frequency</i>	<i>Percentage</i>
<b>0 – 10</b>	03	2.27
<b>11 – 20</b>	15	13.63
<b>21 – 30</b>	11	10.00
<b>31 – 40</b>	19	17.27
<b>41 – 50</b>	41	37.27
<b>51 – 60</b>	16	14.54
<b>&gt; 60</b>	05	4.54
<b>Total</b>	110	100

It was found in the study that n=54 (49.09%) of the participants were illiterate, with farmers having the highest frequency followed by labourers. Trauma and post-operative conditions were risk factors for patients with suspected keratitis. Out of the n=67 conjunctival swabs bacterial isolates were obtained from n=29(43.28%). Out of n=34 Corneal scrapings grew n=3(11.76%) bacterial isolates and n=15(44.11%) fungal isolates and n=1 mixed growth. From the n=28 vitreous fluid samples n=10(35.71%) bacterial and n=5(17.8%) fungal isolates and from lacrimal pus samples bacterial growth was obtained in n=18(75%) of cases.

In vitreous and corneal specimens, growth was observed in n=20 (32.25%) of the n=26 (41.93%) instances that were found to be positive by direct microscopy (KOH mount with Calcofluor White staining), while no growth was found in n=6 cases. Out of n=36 (58.06%) cases that were negative on direct microscopy, growth was seen in n=2 (5.55%) cases, whereas no growth was seen on culture in the remaining n=34(94.44%) cases. Direct microscopy had a sensitivity of 80.12% and a specificity of 79.61%. Out of total n=20 fungal isolates found n=15 cases were from corneal scrapings which yielded *Aspergillus sp.* In n=6, *Penicillium sp.* n=5 *Candida albicans* n=2 and *Fusarium sp.* n=2 and the one mixed growth of candida along with Coagulase negative staphylococcus.

The predominant Gram-negative bacterium isolated was *Pseudomonas aeruginosa* followed by *Klebsiella pneumoniae* and *E. coli*. *P. aeruginosa* showed 100% sensitive to polymyxin B, and 71.5% sensitivity to amikacin and 82.34% sensitive to ceftazidime. Similarly, *K. pneumoniae* showed 100% sensitivity to amikacin and polymyxin B. 91.25% sensitivity to ceftazidime and 82.64% sensitive to gentamycin and 74.36% sensitivity to ofloxacin. The *E. coli* was sensitive in 100% cases to amikacin, and polymyxin B. 91.54% sensitive to ceftazidime, ofloxacin and 80.12% sensitive to gentamycin and least of all 65.74% sensitive to ciprofloxacin.



**Figure 1: Antibiotic sensitivity pattern of Gram-positive isolates**

All the gram-positive isolates were 100 sensitive to vancomycin in addition *S. pneumoniae* was 100 sensitive to Tobramycin, ofloxacin and Erythromycin. However, it was resistant to ampicillin, amoxicillin, and Cefixime. CoNS were 100 sensitive to vancomycin and only 78% sensitive to amikacin and the sensitivity ranged from 50 – 60% in most of the antibiotics and it was only 25% sensitive to ampicillin. In cases of *S. aureus* after vancomycin it was only 70% sensitive to amikacin and the sensitive ranged from 30 – 60% in most of the antibiotics and it was only 29% sensitive to ampicillin the sensitivity pattern is given in figure 1.

## Discussion

Ocular infections are currently thought to be one of the main causes of non-fatal debilitating illnesses in both high-income and low-income nations. <sup>[11]</sup> Up to 5% of all cases of blindness may be a result of an infection brought on by ocular damage. Between 1.5 and 2 million incidents of blindness are attributed to eye infections annually in the world. <sup>[12]</sup> Ocular infections are more common in regions of the world where access to healthcare is limited, health indicators are lower, and a greater number of employees are employed in high-risk professions like farming and agriculture. <sup>[13]</sup> The majority of patients in our study were between the ages of 41-50 years, with a mean age of  $44.58 \pm 11.25$  years and a male to female ratio of 1.7:1. These outcomes agreed with the previous reports. <sup>[14-16]</sup> Our study's male predominance was explained by their outdoor pursuits. Due to the nature of their jobs, agricultural workers and labourers have high incidence rates. Farmers are typically subjected to trauma by some organic materials (such as corn or dry rice stems), which favours fungus invasion. <sup>[17]</sup> Lower understanding of health & hygiene practises and local medical problems, which renders them more susceptible to infections, can be used to explain a higher prevalence of ocular infections in rural populations (52%) and illiterate people (57%). <sup>[18, 19]</sup> Trauma (70%) contact lens use (5%) and post-operative cases were the main predisposing variables for patients with suspected keratitis (8%). In their investigation on the etiological determinants for the identification of microbial keratitis, Hitesh J et al. discovered that endophthalmitis cases were 85.7 percent post-operative, 7.2 percent post-traumatic, and 3.5 percent endogenous. Similar findings were observed by research conducted in South India by Assudani HJ et al., <sup>[20]</sup> Direct microscopy's sensitivity for identifying fungal components in diverse ocular specimens was 80.12%, which

was comparable to Sharma et al.,<sup>[21]</sup> sensitivity study of 81.2%. As a result, direct microscopy performed well in identifying fungal components in our investigation, and its high diagnostic sensitivity, which can be compared to culture, was clearly demonstrated. Furthermore, culturing is a labor-intensive laboratory technique that is rarely used in clinical practice. Predominant isolate identified among positive conjunctival specimens n=29 was *Staphylococcus aureus* 51.72% followed by 20.68% *Coagulase Negative Staphylococci* (CoNS), 6.89 *S. pneumoniae*, *Klebsiella sp.*, *P. aeruginosa* and *E. coli* each. Similar research conducted by Ra'ad et al.,<sup>[22]</sup> and Samuel S O et al.,<sup>[23]</sup> has revealed *Staphylococcus aureus* to be the most common isolate in conjunctivitis. A.O. Okesola et al.,<sup>[24]</sup> also identified *Coagulase-negative Staphylococci* as a secondary common isolate. In a study by Muluye D et al.,<sup>[25]</sup> *Klebsiella pneumoniae* was the most often isolated gram-negative bacterium. 32 The bacterial isolates identified from the positive vitreous samples were CoNS 50% followed by *S. aureus* 30% and 20% *Pseudomonas*. Data from a study by Kodati S et al., which identified *Coagulase-negative Staphylococci* (CoNS) as the most prevalent isolate, are compatible with this data (54.6%). The common fungal isolates found n=15 cases were from corneal scrapings causing keratomycosis found *Aspergillus sp.* In n=6, *Penicillium sp.* n=5 *Candida albicans* n=2 and *Fusarium sp.* n=2 and one case of *candida albicans* with *Coagulase-negative staphylococcus*. Similarly, *Aspergillus sp.* was identified as the most frequently isolated fungus in the Keratomycosis study by Arora U et al.,<sup>[27]</sup> In the current study the Gram-positive isolate among which most frequent was *staphylococcus aureus* was found to be 100% sensitive to vancomycin 70% sensitive to amikacin and the sensitive ranged from 30 – 60% in most of the antibiotics (figure 1). In a similar study by Rajesh S. et al.,<sup>[19]</sup> all Gram-positive isolates were completely responsive to vancomycin, with *Staphylococcus aureus* being most susceptible to aminoglycosides and then fluroquinolones. In our study *S. pneumoniae* was 100 sensitive to Tobramycin, ofloxacin and Erythromycin. Among the gram-negative organism sensitivity was 100% to polymyxin B and ceftazidime and 90% in case of amikacin and 80% in cases of gentamycin and 74% in cases of gentamycin. Similar to the research conducted by Whitcher JP et al.,<sup>[27]</sup> gram negative isolates demonstrated highest sensitivity to Amikacin, ceftazidime, ofloxacin, polymyxin B, and Gentamycin.

## Conclusion

Ocular infections are one of the most prevalent infections in our nation due to subtropical climate. The anterior route directly infects the anterior portion of the eye, whereas blood-borne illnesses have the potential to infect the posterior portion of the eye. Emerging drug resistance is a matter of serious concern and hence all ophthalmological samples must be analysed as thoroughly as possible for antimicrobial resistance due to the increase of antibiotic resistance. Ophthalmologists must adhere to the etiologic approach to diagnosis and consider risk reduction in order to lessen the burden of ocular infections.

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