Microbial profile of the vitreous aspirates in culture proven exogenous endophthalmitis: A 10-year retrospective study

*H Bhattacharjee, K Bhattacharjee, K Gogoi, M Singh, BG Singla, A Yadav

**Abstract**

**Purpose:** To describe the microbiological profile and clinical outcome in the eyes with culture-proven exogenous endophthalmitis. **Methods:** A retrospective analysis of 495 eyes diagnosed as exogenous endophthalmitis was performed over a period of 10 years. In all, aseptically collected aqueous and vitreous aspirates were cultured for bacteria and fungus using standard microbiological techniques. Gram-stain and KOH preparation of the specimens were also performed. The antibiotic susceptibility testing for bacterial isolates was performed by Kirby–Bauer disk diffusion method. The treatment was modified according to the antibiotic sensitivity profile. The final clinical ocular condition was divided into improved, stable or deteriorated. **Results:** Of 148 culture-proven endophthalmitis eyes, 137 (92.57%) were referred from elsewhere, and 11 (7.43%) belonged to our institute. Aetiologically, 76 (51.35%) eyes were post-cataract surgery, 61 (41.22%) were post-traumatic, 5 (3.38%) eyes post-intravitreal anti-vascular endothelial growth factor injection, 5 associated with corneal diseases and 1 bleb-related endophthalmitis. In 31 (20.95%) eyes, primary intravitreal antibiotics were given outside. The cultures revealed monomicrobial growth in 92.57% (n = 137) and polymicrobial growth in 7.43% (n = 11). Among the bacteria (n = 121, 81.76%), *Pseudomonas* species dominated overall (n = 32, 27.11%) and post-operative (n = 26, 38.23%) endophthalmitis group. *Staphylococcus epidermidis* (n = 14, 28%) was prominent in post-traumatic endophthalmitis group. Ninety-two percent (n = 108 isolates) of bacteria were sensitive to vancomycin. In 78 (52.7%) eyes, the clinical ocular condition improved or remained stable while deteriorated in 51 (34.46%). **Conclusion:** A bacterial predominance was observed among causative organisms of exogenous endophthalmitis with *Pseudomonas* species being the most common. The appropriate surgical intervention improved or stabilised the visual acuity in nearly 50% eyes.

**Key words:** Culture positive, exogenous endophthalmitis, outcomes, *Pseudomonas* endophthalmitis

**Introduction**

Infectious endophthalmitis is a vision-threatening and potentially devastating intra-ocular infection caused by an array of organisms. It typically presents with suppurative inflammation of the internal ocular tissues and/or ocular fluids. The intra-ocular entry of infective organisms could be from outside the body, i.e., exogenous or from a systemic (endogenous) infective focus, the later constituting only 2–15% of overall cases.[1] Though endophthalmitis is a rare clinical entity, it can quite possibly lead to devastating outcomes either in the form of permanent blindness, disharmonisation of the eyeball structures (phthisis bulbi) or loss of the eyeball (evisceration), depending on various factors.[2]

Exogenous endophthalmitis can occur after intraocular surgery, penetrating ocular trauma and fulminant infection of adjacent structures. It can have heterogeneous aetiology with variable pattern of the disease course even with treatment and intervention. The sheer number of cataract surgeries performed worldwide, makes it the leading cause (90%) of postoperative endophthalmitis with an incidence of 0.03–0.68%.[2] Several large series estimated the post-traumatic endophthalmitis for much higher incidence, i.e., 3–17%, which is further increased by retained intraocular foreign body.[3]
Microbiologically, the list of offending infective agents is wider in exogenous endophthalmitis, which mainly includes Gram-positive and Gram-negative bacteria, fungi and mixed growth. Gram-positive bacteria and polymicrobial infection are the prominent organisms isolated from post-operative and post-traumatic endophthalmitis eyes, respectively.[2,3] Most reports published Gram-positive coagulase negative staphylococci (Staphylococcus epidermidis) as the leading organism isolated from post-cataract surgery endophthalmitis.[3,4] In post-traumatic endophthalmitis, both environmental and host flora may be implicated which commonly include S. epidermidis, Streptococcus species, Bacillus species and filamentous fungi.[5]

Recently, with the advent of frequent intravitreal injections of anti-vascular-derived endothelial growth factor (VEGF) agents for various ocular disorders, the high theoretical chances of endophthalmitis remain a concern. This leads to the superfluous use of topical antibiotics which can pose a potential challenge for the development of antibiotic resistant bacterial strains which may further reduce the overall success.[6] Oral Streptococcus species have been reported to be the most common for causing post-injection endophthalmitis.[7]

The timely detection and appropriate management of this undesired ocular condition can possibly prevent irreversible vision loss. The empirical use of intravitreal antibiotics have stood the test of time and have provided us with an efficacious management protocol of this acute ocular situation.[2,4,5] The purpose of this study is to highlight the microbiological profile and associated clinical outcome in the eyes with culture exogenous endophthalmitis. As per our knowledge, this is the first study on exogenous endophthalmitis from the North-East part of India, and we describe a detailed microbiological picture along with treatment trends in this part of the country.

Methods

This retrospective study was conducted at Sri Sankaradeva Nethralaya, Guwahati, Assam, which is a Tertiary Eye Care Referral Hospital of the North-East India. Medical records of 495 diagnosed patients having exogenous endophthalmitis presenting from January 2003 to December 2013 (i.e., 10 years) were analysed. The study sample comprised 148 culture positive vitreous aspirates from the affected eyes of 148 patients out of which 137 were referred from elsewhere. As per the standard protocol of our institute, the diagnosis was based on clinical evaluation and B-scan ultrasonography (USG). The clinic files mentioned a thorough record of clinical history, presentation delay, clinical examination, USG, treatment given, microbial spectrum, culture sensitivity and final outcome.

The clinical presentation of post-operative endophthalmitis was classified into fulminant, acute and chronic type depending on the time of occurrence. The fulminant disease was defined as the onset of symptoms within 4 days, the acute within 6 weeks and chronic more than 6 weeks. As per institutional and endophthalmitis vitrectomy study guidelines, the patients were managed based on the visual acuity at presentation. Microscopy and culture sensitivity charts were also reviewed for the analysis. The vitreous specimens were collected via pars plana route either through a needle or vitreous cutter.

As per standard protocol followed at our institute, the intra-ocular specimens-aqueous fluid (AC tap) and vitreous humour were processed within 30 min of their collection. Cultures and smears for detection of bacterial and fungal agents were carried out on the specimen. For culture, the specimens were first inoculated for the culture of bacteria and fungus. The culture media used were 5% sheep blood agar, 5% sheep blood chocolate agar (CA), brain heart infusion broth, thioglycollate broth and Sabouraud dextrose agar (SDA) (Hi-Media, Mumbai). All the inoculated media were incubated at 37°C except SDA, which was incubated at 25°C. CA was incubated in an atmosphere of 10% CO₂ (anaerobic system Mark V Jar, Hi-Media). All the culture media for bacteria and fungus were incubated for 2 weeks in case of no growth before declaring the sample as sterile. The isolated fungi and bacteria were identified by standard methods.[8-10]

Direct smears were prepared from the AC tap and vitreous specimens-one KOH preparation for detecting fungi, a Gram-stain and a Giemsa stain for cytology. Antibiotic susceptibility testing was performed by standard Kirby–Bauer disk diffusion technique. The criteria used to consider the isolated microbial agent as the causative agent were - (i) growth on a single medium correlating with direct smear findings, (ii) growth of the same organism on two or more of the inoculated media and/or (iii) confluent growth in any solid medium. Patient demographics, microbiological profile, follow-up interventions and clinical outcome were assessed.

Results

Of total, 495 exogenous endophthalmitis eyes, only 148 (29.9%) were culture positive and 347 (70.1%) were culture-negative endophthalmitis. Overall, 285 (57.57%) eyes received intravitreal antibiotics before presenting to us, namely, vancomycin + ceftazidime (n = 113, 39.65%), vancomycin + amikacin (n = 97, 34.03%) and only intravitreal vancomycin (n = 75, 26.32%). None, 13 (13.4%) and 18 (24%) eyes were culture positive, respectively. Culture positivity was seen in 117 (55.71%) eyes of 210 eyes which did not receive any intravitreal antibiotics.

Over the 10 years period, 148 culture positive exogenous endophthalmitis eyes from 495 endophthalmitis patients
were identified [Figure 1]. Demographic details, presenting features and aetiology are summarised in Table 1. One hundred and nineteen (80.40%) patients belonged to Assam state. In symptomatology, diminution of vision was complained by 131 (88.51%), ocular pain by 102 (68.92%) and redness by 98 (66.21%) patients. Photophobia, watering and foreign body sensation were complained by 94 (63.51%) of patients. In post-operative endophthalmitis, 15 (19.74%) presented with fulminant disease, 43 (56.58%) had an acute presentation and 18 (23.68%) patients had chronic endophthalmitis.

Before administering any treatment, B-scan USG was performed in all patients. Vitreous echoes suggestive of inflammation were present in all while none had retinal detachment. Table 2 highlights the treatment administered to the patients. Seventy (47.29%) patients underwent pars plana vitrectomy with intravitreal antibiotics while 29.05% were subjected to vitreous tap with intravitreal antibiotics. Additional procedures such as globe repair, intraocular foreign body removal, lensectomy, intraocular lens explanation and AC wash with antibiotics were performed as required. Evisceration was required in 8 (5.40%) eyes.

The microbiological profiles of the cultured specimens are shown in Table 3a and b [Figure 2]. Pure cultures of a single bacterium were found in 121 (81.76%), fungi in 16 (10.81%) and more than one organism in 11 (7.43%) specimens. The causative agents in fulminant cases were mainly Gram-negative organisms 11 (73.33%) while Gram-positive bacteria 38 (88.37%) were predominant in acute cases. The majority (n = 9, 50%) of fungi were isolated from the chronic endophthalmitis group. Of 5 post-intravitreal anti-VEGF injection, 4 (80%) were positive for *Bacillus cereus* while *S. epidermidis* was isolated from 1 vitreous aspirate. All patients with positive fungal culture received intravitreal amphotericin-B (10 μg/0.1 ml) which was repeated as required clinically.

### Table 1: Demographic profile and aetiology of culture proven exogenous endophthalmitis patients

<table>
<thead>
<tr>
<th>Patient profile</th>
<th>Number of patients (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total culture positive eyes (%)</td>
<td>148 (29.9)</td>
</tr>
<tr>
<td>Gender (%)</td>
<td></td>
</tr>
<tr>
<td>Males</td>
<td>99 (66.89)</td>
</tr>
<tr>
<td>Females</td>
<td>49 (33.10)</td>
</tr>
<tr>
<td>Referral pattern (%)</td>
<td></td>
</tr>
<tr>
<td>Outside</td>
<td>137 (92.62)</td>
</tr>
<tr>
<td>Our institute</td>
<td>11 (7.38)</td>
</tr>
<tr>
<td>Mean age (years)</td>
<td>42.64 (1-82)</td>
</tr>
<tr>
<td>Average symptom to surgery interval (median - 1.5 days)</td>
<td></td>
</tr>
</tbody>
</table>

### Table 2: Treatment administered on presentation

<table>
<thead>
<tr>
<th>Treatment modality</th>
<th>Number of cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pars plana vitrectomy + IV antibiotics (+IOLF removal, lensectomy/IOL explantation, AC wash)</td>
<td>70</td>
</tr>
<tr>
<td>Vitreous tap/biopsy + IV antibiotics (+globe repair, FB removal, AC wash)</td>
<td>43</td>
</tr>
<tr>
<td>Multiple procedures in repeated steps</td>
<td>26</td>
</tr>
<tr>
<td>Evisceration ± ball implant</td>
<td>8</td>
</tr>
<tr>
<td>No records</td>
<td>1</td>
</tr>
<tr>
<td>Total</td>
<td>148</td>
</tr>
</tbody>
</table>

*285 (57.57%) patients received intravitreal antibiotics before presenting to our institute

### Table 3a: Aetiological distribution of the cultured organisms in post-operative and post-traumatic endophthalmitis group

<table>
<thead>
<tr>
<th>Organism</th>
<th>Number of cultures</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cataract surgery</td>
<td>76</td>
</tr>
<tr>
<td>Ocular trauma</td>
<td>61</td>
</tr>
<tr>
<td>Intravitreal injections</td>
<td>5</td>
</tr>
<tr>
<td>Penetrating keratoplasty</td>
<td>2</td>
</tr>
<tr>
<td>Infectious keratitis</td>
<td>2</td>
</tr>
<tr>
<td>Bleb-related</td>
<td>1</td>
</tr>
<tr>
<td>Infected lipid keratopathy</td>
<td>1</td>
</tr>
</tbody>
</table>

*Figure 1: Pie diagram showing number of patient and distribution

*Figure 2: Aetiological distribution of the cultured organisms in post-operative and post-traumatic endophthalmitis group

www.ijmm.org
Antibiotic sensitivity was performed for the commonly used intravitreal and topical antibiotics using Kirby–Bauer disk diffusion method in all. For Gram-positive aerobic bacteria-vancomycin, ofloxacin, gatifloxacin, moxifloxacin, cefturoxime and ceftriaxone were tested. For Gram-negative, bacterial-vancomycin, ofloxacin, gatifloxacin, moxifloxacin, amikacin and cefotaxime were tested. For Gram-negative, testing for ceftazidime, ciprofloxacin, amikacin and tobramycin was conducted. Sixty-six (54.55%) isolated bacterial colonies showed moderate to high sensitivity to amikacin and 55 (45.45%) to cefotaxime (33.3%). Sensitivity to ceftazidime, ciprofloxacin and tobramycin was seen for 67 (55.37%) and 59 (48.76%), respectively. All the Gram-positive bacteria were sensitive to vancomycin.

The clinical outcome for various causative organisms is mentioned in Table 4. Post anti-VEGF injection endophthalmitis eyes deteriorated rapidly to no light perception status. Clinical improvement in visual acuity was noticed in 53 (35.81%) patients, while 25 (16.89%) remained stable. Fifty-one (34.46%) eyes deteriorated even after maximum advisable surgical intervention. Nineteen (12.84%) patients were lost to follow-up.

Discussion

This is the first study of its kind from The Northeast part of India mentioning the microbiological profile and clinical outcome in eyes managed for exogenous endophthalmitis. The organisms causing this devastating infection vary according to the regions and climatic conditions all over the world hence making the findings of this study important. In India, fungi cause 10–20% of post-operative endophthalmitis, while the dominance of bacteria is undoubted in west.[7-15] Fungal infection is associated with poorer prognosis and indolent course of the disease.[13-15]

The culture positive rate of our study was 29.89% (n = 148 eyes) which is less as compared to other reports from India by Lalitha et al. (53%) and Gupta et al. (52.5%).[12,14] This can be attributed to the reason that 57.57% eyes received intravitreal antibiotics elsewhere, before presentation to us. This might have reduced the load of the viable microbes in the inoculums.[5]

The bacteria isolated in post-operative endophthalmitis generally are same as that of same individual’s oropharyngeal and mucosal flora.[16] Gram-positive bacteria mainly coagulase-negative Staphylococcus (CoNS) are predominant (65.64%) in acute variety of endophthalmitis. The Staphylococcus aureus and Streptococcus viridians follow as 9.9% and 9%, respectively.[3,16] In contrast, our study showed 22.88% of CoNS isolation out of the bacterial colonies which are considerably different from the literature.
The highlighting feature of our study was the isolation of large (26.45%) number of *Pseudomonas* species out of the 121 bacterial colonies. This is in contrast to 11.53% as reported by a South Indian study. As observed over various studies, the common cause of organism entry into the eye is secondary to infected irrigating solutions, inadequately sterilised instruments and the operative environment. The isolation of *Pseudomonas* and *Aspergillus* species has been found to be significantly high in hot and humid climates. Theoretical chances of cluster endophthalmitis by these virulent bacteria in the mentioned predisposing environment can lead to mass level disaster. This is common in a makeshift surgical camp arrangement scenario.

Exogenous endophthalmitis may follow any intravitreal injection with a possibility of an increase in its incidence secondary to the expanding indications and frequency of intravitreal anti-VEGF therapy. We experienced a cluster of five cases from our institute following the intravitreal anti-VEGF injection given on a single day, of which four eyes were positive for *B. cereus*. However, a Canadian study reported oral *Streptococcus* as a most common causative organism for post-intravitreal anti-VEGF injection endophthalmitis.

Our study shows that despite receiving intravitreal isolated vancomycin elsewhere, the culture was positive in 24% of eyes, which is in contrast to the Kirby–Bauer sensitivity of isolates to vancomycin (>92%). This can be attributed to the improper dispensing, dosage and method of injection of the drug. The final ocular condition either improved or remained stable in 52.7% eyes after maximum possible interventions while 34.46% deteriorated. This can be ascribed to the delayed presentation of the patients to our tertiary care referral centre. The final status of the 19 (12.83%) eyes was not known as patients were lost on follow-up. The limitations of our study are its retrospective nature and non-use of PCR (polymerase chain reaction) technology for diagnosis which could have increased the organism isolation rate for our study.

**Conclusion**

The authors want to stress on the necessity to strictly follow and adhere to the standard surgical protocols and endophthalmitis prophylaxis guidelines before performing intraocular surgeries. This possibly can bring down the rate of exogenous endophthalmitis in post-operative patients, specifically *Pseudomonas*-related nosocomial infections. In addition to this, the standard dosage, dispensing and method of delivering intravitreal antibiotics, while managing endophthalmitis, should also be followed according to the standard guidelines. Religious adherence to the standardised protocols can prevent and provide better treatment for this dreaded intraocular complication. To increase the recovery rate of causative microorganism, the ideal method of vitreous specimen collection, inoculation and modern technology like PCR can play a major role.

**Acknowledgement**

The authors thank Dr. Satyan Deka, Dr. Manab Jyoti Barman, Dr. Ronel Soibem for their support.

**Financial support and sponsorship**

Sri Kanchi Sankara Health and Educational Foundation, Beltola, Guwahati - 781 028, Assam, India.
Conflicts of interest

There are no conflicts of interest.

References