



ETIOLOGICAL AGENTS AND THEIR SUSCEPTIBILITY TO VARIOUS DRUGS IN CASES OF ENDOPHTHALMITIS

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ABSTRACT

PURPOSE: To find out the various bacteriological agents responsible for endophthalmitis and determine their susceptibility to antibiotics in the patients of a tertiary healthcare centre in Gwalior.

METHODS: Fifty cases of endophthalmitis were studied at a tertiary healthcare centre in Gwalior. Relevant clinical history was obtained. Vitreous tap were obtained and was send for microbiological evaluation to determine the etiologic agent and antibiotic sensitivity.

RESULT: Positive growth was found in 74% of cases. Seven different types of bacteria were isolated. Gram-positive (86.38%) bacteria were isolated > Gram-negative (13.62%) bacteria. Single growth (70.45%) > mixed growth (29.54%). Coagulase negative staphylococci epidermidis (34.09%) were most commonly isolated, followed by staphylococcal aureus (20.45%), streptococcus pneumonia (18.18%), streptococcus pyogenes (6.81%), streptococcus viridians (6.81%), E.coli (6.81%), Hemophilus influenza (6.81%). In the present study out of 38 Gram-positive isolates vancomycin is the most sensitive drug (100%), followed by Gentmycin (94.73%), ciprofloxacin and ofloxacin (86.84%), cephalixin (84.21%), cephalzoline (73.68%), chloramphenicol (65.78%). Tobramycin was the least sensitive (52.63%).

CONCLUSION: The most common bacterial isolate prevailing in the patients of endophthalmitis in this geographical area was Staphylococcus Epidermidis (Gram-positive, coagulase negative). During study about antibiotic susceptibility, most of the Gram positive isolates were sensitive to antibiotic Vancomycin and the Gram-negative isolates were sensitive to antibiotic Ceftazidime and Amikacin

KEYWORDS : Endophthalmitis, Antibiotic Sensitivity, Vitrectomy

INTRODUCTION

Endophthalmitis is a devastating inflammatory process that can lead to blindness. In endophthalmitis there is suppurative inflammation of vitreous cavity along with retinal and uveal component of the eye¹. In vast majority of cases of endophthalmitis the inflammation is triggered by infectious agent². The source of such infectious agent could be an exogenous such as following trauma or after surgery. The infectious agent encountered following the eye surgery or trauma is usually the organism harboring the outer surface of the eye^{2,3}. Endogenous endophthalmitis is less common and occur secondary to hematogenous dissemination and spread from distant infective source in the body^{2,3,4,5}.

In India gram positive bacteria may account for 53% of the post operative cases of endophthalmitis, 26% may be due to gram negative bacteria rest 17% due to fungal infection^{6,7}. Recently endophthalmitis is a major reason for evisceration⁸.

According to the endophthalmitis vitrectomy study; postoperative endophthalmitis is divided generally in to two types: acute and chronic. Acute postoperative endophthalmitis is defined as infections within 6 weeks of surgery; on the other hand chronic postoperative endophthalmitis is defined as infections after 6 weeks of surgery⁹.

The term chronic postoperative endophthalmitis (CPE) was first coined in 1986 in a case series of 15 patients by Meisler et al¹⁰. The inflammation is usually indolent and may persist for months. It is often misdiagnosed as non infectious iritis where it improves initially with topical corticosteroid therapy while flaring whenever corticosteroids are tapered or stopped¹¹.

This is in contrast to acute postoperative endophthalmitis which presents as a single episode of severe inflammation with an acute onset that usually follows surgery by a few days but can be delayed more than a week in some cases^{10,11}.

AIMS AND OBJECTIVES

To find out the various bacteriological agents responsible for endophthalmitis and determine their susceptibility to antibiotics in the patients of JA Group of hospitals.

MATERIAL AND METHODS

This study will be conducted on 50 eyes of endophthalmitis patients in the Department of Ophthalmology and Department of Microbiology G. R. Medical collage Gwalior from February 2015 to August 2016.

The case will be recorded in a predesigned proforma including detailed history and relevant examination including local ocular examination, anterior chamber tapping, posterior chamber tapping for culture and sensitivity.

Samples were collected from all the cases under complete aseptic precautions and were subjected to culture and sensitivity.

In all cases routine diagnostic test were done on the first day. On the same day vitreous tap was collected for bacteriological examination. Special care was taken not to use any chemotherapeutic or antibiotic agent before collecting the specimen for bacteriological study.

SELECTION CRITERIA:

1. Patients with age > 15 years < 80 years.
2. Already diagnosed and willing to sign an informed consent and able to comply with the requirement of the study.
3. Postoperative endophthalmitis patients.
4. Endogenous endophthalmitis patients.
5. Traumatic endophthalmitis patients.

EXCLUSION CRITERIA

1. Refusal to participate in the study.
2. Inability to follow protocol.
3. Patients with age < 15 years > 80 years.

METHOD

All the patients underwent careful history taking and relevant ocular examination was done.

Procedure for obtaining vitreous tap:

- Povidone-iodine 5% solution poured in eye.
- Surgical drape, lid speculum is applied.
- 30g needle attached to TB syringe inserted at limbus for A/C tap, without collapsing A/C.
- Obtain 0.1 cc.
- Vitreous specimen, obtain by either vitreous needle tap or by

vitreous biopsy with a cutting/aspirating probe.

- Vitreous Needle tap:
- Vitreous needle tap is performed by inserting 27 to 22 g needle attached to a TB syringe in to the vitreous cavity through the pars plana.
- Slowly aspirate, if no fluid vitreous can be obtained with a needle tap, a vitreous biopsy must be performed instead to avoid aspirating formed vitreous.

The various culture media used were-

- 1) Nutrient agar media – for primary culture.
- 2) Mac-Conkey agar – this was used to differentiate the lactose fermenting and non-lactose fermenting gram-negative bacilli.
- 3) Blood agar media – to study fastidious bacteria/ hemolytic bacteria.
- 4) Peptone water – for isolation of pure culture and doing the antibiotic sensitivity test.
- 5) Sugar media –for biochemical reaction for acid and gas production.

Identification of organisms:

- Isolated colonies from plates were picked up by a loop and inoculated in peptone water media and incubated for 1-2 hours at 37°C.
- The motility of the organisms grown was seen by hanging drop preparation.
- Further identification was done by various biochemical tests.
- The following tests were performed to determined the specific characters of the organisms-

1. Catalase test :
2. Coagulase test:
3. Oxidase test:
4. Indole reaction
5. Methyl Red test:
6. Voges-proskauer test:
7. Citrate utilization:
8. Urease test:
9. Sugar fermentation:
10. H₂S production:
11. Bacitracin disc sensitivity test:
12. Optochin disc sensitivity test:
13. Bile esculin test:

Antibiotic sensitivity test:

The antibiotic susceptibility pattern was done by kirby-Baur disc diffusion method. Antibiotic discs were placed aseptically on a muller-Hinton agar plate swabbed with test bacteria pre grown to 0.5 Macfarland standards in nutrient broth. The plate was then incubated overnight at 37°C for 24 hours. The zone of inhibition (ZOI) around the disc was measured with a ruler and compared to standard interpretation chart. The quality of each test was maintained by using standard procedures. The quality of each agar plate prepared was ensured by incubating one plate of each lot in the incubator control strains of E.coli (ATCC 25922) and staphylococcus aureus (ATCC 25923) were used for the standardization of the Kirby-Bauer test and also for correct interpretation of the zone of diameter. The quality of the sensitivity test was maintained by maintaining the thickness of MHA at 4 mm and the PH at 7.2-7.4. Strict aseptic conditions were maintained while carrying out all the procedure.

OBSERVATIONS AND RESULTS:

Table 1: Etiological factors in endophthalmitis

S. No.	Etiological Factor	No. Of Cases	Percentages
1	Post-operative	35	70%
2	Post-traumatic	10	20%
3	Endogenous	5	10%
4	Others	0	0%

Table 2 : Showing the bacteriological examination of vitreous fluid tap in endophthalmitis

S. NO.	Organism	No. of cases	%
1	Staphylococcus epidermidis	14	37.6
2	Staphylococcus aureus	6	16.21
3	Streptococcus pneumonia	6	16.21
4	Streptococcus pyogens	2	5.40
5	Streptococcus viridians	1	5.40
6	E.coli	1	2.70

7	H.influenzae	1	2.70
8	Staph.aureus+Strep. pneumonia	1	2.70
9	Staph.aureus mixed with E.coli	1	2.70
10	Staph. aureus+ H.influenza	1	2.70
11	Strep.pneumoniae, H.influenza+strep pyogenes	1	2.70
12	Staph.epidermidis+Strep pyogenes	1	2.70
13	E.coli +Strep viridians	1	2.70
	TOTAL	37	100

Table 3 : Pattern of organism isolated from single and mixed growth

S. NO	Organism isolated in growth	No. of organism in single growth	No. of organism in mixed growth	TOTAL [Positive
1	CONS[Staph. epidermidis]	14[45.16%]	1[7.69%]	15[34.09%]
2	Staphylococcus aureus	6[19.35%]	3[23.07%]	9[20.45%]
3	Streptococcus pneumonia	6[19.35%]	2[15.38%]	8[18.18%]
4	Streptococcus pyogenes	1[3.22%]	2[15.36%]	8[18.18%]
5	Streptococcus viridians	2[6.45%]	1[7.69%]	3[6.81%]
6	Escherichia coli	1[3.22%]	2[15.36%]	3[6.81%]
7	Hemophilus influenza	1[3.22%]	2[15.36%]	3[6.81%]
	TOTAL	31[70.45%]	13[29.54%]	44[100%]

Table 4 : Antibiotic sensitivity pattern among gram positive isolates

S. NO.	Antibiotics	Susceptibility Of Microorganisms				
		CONS [Staphylococcus epidermidis] (%)	Staphylococcus aureus (%)	Streptococcus pneumoniae (%)	Streptococcus pyogenes (%)	Streptococcus viridians (%)
1	Vancomycin	15[100%]	9[100%]	8[100%]	3[100%]	3[100%]
2	Chloramphenicol	12[80%]	4[44.44%]	5[62.5%]	2[66.66%]	2[66.66%]
3	Gentamycin	15[100%]	8[88.88%]	7[87.5%]	3[100%]	3[100%]
4	Ciprofloxacin	13[66.66%]	8[88.88%]	7[87.5%]	2[66.66%]	3[100%]
5	Ofloxacin	13[86.66%]	8[88.88%]	7[87.5%]	2[66.66%]	3[100%]
6	Cephazolin	12[80%]	7[87.5%]	5[62.5%]	2[66.66%]	2[66.66%]
7	Cephalexin	13[86.66%]	8[88.88%]	6[75%]	3[100%]	2[66.66%]
8	Tobramycin	8[53.33%]	4[44.44%]	4[50%]	2[66.66%]	2[66.66%]

Table 5: Antibiotic sensitivity patterns among gram negative isolates

S. NO.	ANTIBIOTICS	Susceptibility Of Microorganism		TOTAL
		E. Coli	H.Influenzae	
1	Ceftazidime	3[100%]	2[66.66%]	5[83.3%]
2	Amikacin	2[66.66%]	2[66.66%]	4[66.66%]
3	Chloramphenicol	2[66.66%]	1[33.33%]	3[50%]
4	Gentamycin	1[33.33%]	2[66.66%]	3[50%]
5	Ciprofloxacin	1[33.33%]	2[66.66%]	3[50%]
6	Ofloxacin	1[33.33%]	1[33.33%]	2[33.33%]
7	Cephazolin	2[66.66%]	1[33.33%]	3[50%]
8	Cephalexin	1[33.33%]	2[66.66%]	3[50%]
9	Nalidixic acid	2[66.66%]	1[33.33%]	3[50%]

DISCUSSION :

Endophthalmitis is one of the most devastating complications after ocular surgery or trauma and in people with systemic infection .Treatment of endophthalmitis remain challenging. Early diagnosis and treatment are essential to optimize visual outcome. Intravitreal

antimicrobial drug application achieves the higher intraocular substance level needed for effective endophthalmitis treatment.

A more detailed understanding of the interactions between offending organisms and the intraocular host response is needed to more effectively treat endophthalmitis to improve visual outcome. Clinical evidence shows that while antibiotics effectively kill intraocular organisms and anti-inflammatory agents suppress the intraocular inflammatory response, these drugs have no effect on the toxins or inflammation-derived enzymes that directly affect retinal function or architecture.

As more information becomes available with respect to the natural course of different types of endophthalmitis, several steps in the evolution of infection may emerge as new therapeutic opportunities.

For the treatment of exogenous endophthalmitis, intravitreal antibiotics need not to be supplemented with intravenous antibiotics. In contrast, most cases of endogenous endophthalmitis, where the primary focus of infection is outside the eye, require systemic antimicrobial therapy.

Age incidence

In the present study, it was observed that the maximum number of cases of endophthalmitis was found to be commonest (72%) between 50 and 70 years of age.

A clinico-microbiological analysis by Savitri Sharma, Tapas R. Padhi, Sarita Kar, Arvind Roy & Tarprasad Das in 2012 showed the mean age of patients with post-operative endophthalmitis is 51.8 year was higher than that of patients with post-traumatic (22.1yr) and endogenous endophthalmitis (30.9yr). However the difference was not significant⁶².

Sex incidence

In the present study males were most commonly affected with endophthalmitis (74%) than females (26%).

Male predominance in postoperative group could be due to greater chances of exposure of males to trauma due to outdoor activities, the reason for male preponderance in other two groups could possibly be due to more males willing to travel when referred or could be socio-economic.

The difference was not significant.

Socio-economic status

In the present study the cases were classified on the basis of per capita income per month according to social classification by P. Kumar (updated by 2001 C.P.I).

Average 64% of the cases of endophthalmitis belong to lower-lower class, and none of the cases belong to upper class. Poverty (lack of means), illiteracy and consequent poor hygiene may be the reason for these findings.

Etiology

1. Postoperative endophthalmitis :

Romero CF, Rai MK, Lowder CY in 1999, Ng EWM, D Amico DJ in 2000 have reported that postoperative endophthalmitis responsible for 72% of endophthalmitis cases¹⁸.

Lemley and Han (2007) reported that 90% of endophthalmitis cases occurring following cataract surgery⁶⁷.

O' Brien et al (2007) find that the rate of occurrence of post cataract endophthalmitis remains low, ranging between 0.06% to 0.25%. Among the type of cataract surgery performed, phacoemulsification accounted for 48% of the postoperative endophthalmitis cases, while 38.5% and 6.6% of cases followed extracapsular and intracapsular extraction⁶⁸.

Endophthalmitis vitrectomy study (EVS-1995) also reported that 90% of endophthalmitis cases develop after cataract surgery²⁰.

In the present study postoperative endophthalmitis is responsible for 70% cases of endophthalmitis.

2. Posttraumatic endophthalmitis :

Zhang Y, Zhang MN, Jiang CH et al in 2010 reported that

posttraumatic endophthalmitis occurred in 11.1% of open globe injury³⁸.

Boldt HC, Pulido JS, Blodi CF et al in 1989 found that the rate of infection in post-trauma cases is much higher (3%-17%). In rural areas the rate of post trauma infections is much higher, approximately 30%⁶⁹. Meredith et al 1999, O' Brien et al 1995 reported that in cases of penetrating injury, 3%-17% of patients develop microbial endophthalmitis⁷⁰.

In the present study posttraumatic endophthalmitis is responsible for 20% cases of endophthalmitis cases.

3. Endogenous endophthalmitis :

Arevalo et al 2010, Pulifito et al 1982 reported that endogenous endophthalmitis accounts for 2%-15% of all cases of infectious endophthalmitis⁴⁹.

Romero CF, Rai MK, Lowder CY in 1999 reported that endogenous endophthalmitis responsible for 2%-8% of infectious endophthalmitis¹⁸.

Chee SP, Jap A in 2001, Shrader SK, Band JD, Murphy P in 1990 find that only 5%-10% of endophthalmitis cases are of endogenous origin⁷¹. In the present study endogenous endophthalmitis is responsible for 10% of endophthalmitis cases.

Clinical features

Endophthalmitis vitrectomy study group in 1995 blurred vision reported in 94.3% patients, 82.1% patients having red eye, 74% patients have pain, 34.5% patients having complain of swollen lid. Hypopyon reported in 85% patients, 79% patients having hazy media²⁰.

In the present study 90% patients reported blurred vision, 80% patients having red eye, 74% patients having pain, swollen lid present in 36% patients. Hypopyon reported in 80% patients, 76% patients having hazy media and yellow reflex seen in 100% patients.

Bacteriology:

Benz MS et al in 2004 in a retrospective study evaluated 278 patients, including all sub-categories of endophthalmitis, and most commonly yielded staphylococcal epidermidis (27%), Gram-positive organism comprised 78.5% of those identified, Gram-negative 11.8%, and fungal 8.6%⁶¹.

According to EVS in 1995, 94.2% of culture positive endophthalmitis cases involved Gram-positive bacteria; 70% of isolates were Gram-positive, coagulase negative staphylococci, 9.9% were staphylococcus aureus, 9% were streptococcus species, 2% were Enterococcus species, 3% were other Gram-positive species. Gram-negative species were involved in 5.9% of cases²⁰.

M.L. Durand in 2013 find that the most common organism responsible for endophthalmitis are coagulase negative staphylococci (70%), staphylococci aureus (10%), streptococci (9%), other Gram-positive cocci including enterococci and mixed bacteria (5%), and Gram-negative bacilli (6%)⁷².

In the present study:

- Positive growth was found in 74% of cases.
- Seven different types of bacteria were isolated.
- Gram-positive (86.38%) bacteria were isolated > Gram-negative (13.62%) bacteria.
- Single growth (70.45%) > mixed growth (29.54%).
- Coagulase negative staphylococci epidermidis (34.09%) were most commonly isolated, followed by staphylococcal aureus (20.45%), streptococcus pneumonia (18.18%), streptococcus pyogenes (6.81%), streptococcus viridians (6.81%), E.coli (6.81%), Hemophilus influenza (6.81%).
- Staphylococcus aureus were most predominant bacteria in mixed growth.

Antibacterial susceptibility pattern:

According to Benz MS et al in 2004 sensitivity of Gram-positive bacteria to Vancomycin is 100%, followed by Gentamycin (78.4%), Ciprofloxacin (68.3%), Ceftazidime (63.6%), and Cephazoline (58%). The sensitivity of Gram-negative bacteria was 94.2% for ciprofloxacin, 80.9% for Amikacin, 80% for ceftazidime and 75% for Gentamycin⁶¹.

According to Recchia FM in a study in 2005 found that sensitivity of Gram-positive organisms for Vancomycin is 99%⁷³.

Anand AR et al in a study in 2000 found that in India sensitivity of Gram-negative organism to amikacin or ceftazidime is only 68% and 63%, respectively⁷⁴.

EVS (Endophthalmitis vitrectomy study) in 2005 showed that only 89.5% of Gram-negative isolates were sensitive to amikacin or ceftazidime²⁰.

In the present study out of 38 Gram-positive isolates vancomycin is the most sensitive drug (100%), followed by Gentmycin (94.73%), ciprofloxacin and ofloxacin (86.84%), cephalexin (84.21%), cephalazone (73.68%), chloramphenicol (65.78%). Tobramycin was the least sensitive (52.63%).

Out of 6 (13.62%) Gram-negative isolates ceftazidime was the most sensitive (83.3%) followed by amikacin(66.66%), Gentamycin, chloramphenicol, cephalazone,cephalexin, Nalidixic acid. Ciprofloxacin and ofloxacin was the least sensitive to Gram-negative isolates.

CONCLUSION :

In the present study, it can be concluded that:

- 1) The most common bacterial isolate prevailing in the patients of endophthalmitis in this geographical area are the Staphylococcus Epidermidis (Gram-positive, coagulase negative).
- 2) During study about antibiotic susceptibility, most of the Gram positive isolates were sensitive to antibiotic Vancomycin and the Gram-negative isolates were sensitive to antibiotic Ceftazidime and Amikacin.

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