Incidence and Sensitivity Pattern of *Pseudomonas aeruginosa* in Chronic Suppurative Otitis Media in South Indian Rural Population

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ABSTRACT

To identify the common aetiology of Chronic suppurative otitis media (CSOM) and to assess the sensitivity pattern of that organism. The ear discharge from 93 patients with a clinical diagnosis of CSOM was collected and was subjected to direct smear and aerobic culture and sensitivity. 93 samples were positive on direct smear or culture. *Pseudomonas aeruginosa* was the most common isolated aerobic organism (40). It was most sensitive to Amikacin (37/40) and least sensitive to Magnex (13/40). *Pseudomonas aeruginosa* was the most common isolate – the sensitivity pattern of the aerobes was tested – which helped in accurate treatment of the condition.

Keywords:- CSOM, *Pseudomonas aeruginosa*

INTRODUCTION

Chronic suppurative otitis media (CSOM) is characterised by a long standing infection of the middle ear commonly seen among infants and children in whom signs of infection are missing. It is characterised by painless discharge dating from months to years and is the single major cause of deafness in India. It is especially common amongst children of low socio economic strata. [1]

The introduction of antibiotics has reduced the incidence of intracranial complications in otitis media. However the occurrence should not be underestimated due to the high associated morbidity and mortality.

In this study, an effort has been made to
a. culture and study the aerobic flora involved in causing CSOM
b. Identify the most common aerobic bacterial aetiology of CSOM
c. Determine the antibiotic sensitivity of the isolated organism.

*Pseudomonas aeruginosa* is the most predominant organism among the cases of CSOM reported by several workers in India and abroad with an incidence ranging from 21% to 52.94%. Gulatietal[2], Brook & Finegold[3], Ayyagiri etal[4], Lalita, Rekharao and Bhaskaran[5], Maninder pal singh et al[6]. A Nandy, PS Mallaya S K Sivaranjan[7] Atluntas A et al[8] and camposetal[9].

MATERIAL AND METHODS

The study group included 93 patients with the clinical diagnosis of CSOM from the department of Otorhinolaryngology, Narayana Hospital, Narayana medical college , Nellore, Andhra Pradesh , India. The ear discharge was collected under aseptic precautions with the aid of an aural speculum, prior to the instillation of any topical medication. The first swab was used to make a smear on a clean glass slide for direct smear examination by Gram’s stain. The second swab was processed for the isolation of aerobic bacteria.
Direct Smear Examination:
Gram’s stain was performed by Jensen’s modification and was then screened under the oil immersion to note the various morphological types of bacteria, their number, the presence or absence of inflammatory cells.

Aerobic Culture:
The swab on reaching the laboratory was inoculated on the following culture media.
1. Mac Conkey agar plate
2. Blood agar plate
3. Chocolate agar plate and nutrient agar plate to isolate the organisms. The inoculated Blood agar and Mac conkey agar plates were incubated aerobically at 37°C for 24 hours.

After overnight incubation at 37 degrees C the blood agar and Mac conkey agar plates were examined for evidence of growth. The colony characters were studied; smears were stained by Gram’s stain and examined under the 100x objective.
After 48 hours incubation the chocolate agar plate was similarly examined and the colonies further processed. The bacterial species than isolated were identified by morphology, cultural characteristics and bio-chemical reactions according to the standard techniques [10]

The Gram negative bacilli were tested for motility by hanging drop and then they were subjected to other biochemical and sugar fermentation test. The tests were read after incubation at 37°C at the end of 24 hours and 48 hours.

Pseudomonas species appeared as pale coloured colonies on Mac conkey agar, some strains produced a greenish blue pigment on nutrient agar. All the species were motile, gave positive results with the oxidase reagent and were catalase positive. Glucose was oxidized to form acid only. All the other sugars and Indole, MR, & VP test were negative. The sugars were used to study the fermentation reaction with Glucose, lactose, sucrose, maltose and Mannitol. The Bio-chemical tests done were Indole, Methyl red, Voges – Proskauer, citrate (IMVIC) and Urease production as per the standard norms [10]

All sugar fermentation and biochemical tests were done from the subcultures made from isolated colonies picked from the primary isolation media. All subcultures were incubated before putting up the sugar and biochemical test. After the subcultures reached MC Farland’s Grade 3 turbidity the sugars and biochemical tests were put up and incubated at 37°C for 24 hours and 48 hours.

Antibiotic Sensitivity Tests
Antibiotic sensitivity testing was done by KirbyBauer disk diffusion method.

The antibiotics used were
- Amoxicillin
- Cephexime
- Cephtazidine
- Ciprofloxacin
- Chloramphenicol
- Amoxiclav
- Cefaperazone with Sulbactum
- Cefataxime
- Sporflaxcin
- Ampicillin
- Erythromycin
- Vancomycin
- Penicillin
- Oxacillin
- Cephaxiline
- Azithromycin

Mueller – Hintor is the standard agar base medium for testing most bacterial organisms, with certain supplements and substitutions again required for testing more fastidious organisms. In addition to factors such as pH and cation content, the depth of the agar medium can also affect test accuracy and must be carefully controlled. Because antimicrobial agents diffuse in all direction from the surface of the agar plate the thickness of the agar affects the antimicrobial drug concentration gradient.
If the agar is too thick, zone sizes would be smaller, if too thin the inhibition zones would be larger.

Inoculation and Incubation:

Before disk placement, the plate surface was inoculated using a swab that had been submerged in a bacterial suspension standardized to match the turbidity of the 0.5 McFarland standard. The surface of the plate was swabbed in three directions to ensure an even and complete distribution of the inoculum over the entire plate. Within 15 minutes of inoculation, the antimicrobial agent disks were applied and the plates were inverted for incubation, to avoid accumulation of moisture on the agar surface which could interfere with interpretation of test results.

For most organisms, incubation was done at 35°C in air, but increased CO₂ was used while testing fastidious bacteria.

RESULTS

The Results of Direct smear and culture:
This table (Table 1) compares the results of direct smears and culture of the specimens. Out of the 93 specimens, 93 (100%) were direct smear positive and culture positive

<table>
<thead>
<tr>
<th>Details of Isolation</th>
<th>Number</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive cultures</td>
<td>93</td>
<td>100%</td>
</tr>
<tr>
<td>Negative cultures</td>
<td>0</td>
<td>0%</td>
</tr>
</tbody>
</table>

Aerobic Bacterial flora isolates:

This chart (Chart 1) shows the incidence of various aerobes isolated from 93 specimens. *Pseudomonas aeruginosa* was the most predominant species in 40 cases (43.01%), followed by *Staphylococcus aureus* (31 cases/33.33%), *Eschericia coli* was present in 12 cases (12.9%) and *Proteus sp* and *Klebsiella sp* were seen in 5 cases each (5.38%)
The bacteriologic study of otitis media revealed the isolation of a variety of organisms. 21 strains were sensitive to gentamycin, 37 strains were sensitive to amoxiclav, 21 strains were sensitive to azithromycin, 21 strains were sensitive to gentamycin, 14 strains were sensitive to ciprofloxacin, 14 strains were sensitive to netilmicin, 14 strains were sensitive to erythromycin, 13 strains were sensitive to magnez.

**DISCUSSION**

According to Senturia (1980) the acute phase of otitis media is considered to be the initial three weeks of inflammation, chronic phase three months following the onset of inflammation and subacute phase is said to be between three weeks and three months of inflammation. [11]

Many studies have investigated the bacterial flora in CSOM. The commonest organisms isolated are *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Proteus*—less common organisms are *E.coli* & Klebsiella species. [11] In this particular study of ninety three cases, all the patients had symptoms lasting more than three months, mainly the symptoms being, ear discharge, pain in the ear and other signs of inflammation like fever. Hence all the cases belong to the group of CSOM.

In the study done, bacteria were demonstrated in direct smears in 100% of specimens and culture could isolate bacteria in 100% of specimens.

Smear Positive  
Culture Positive  
\[93\]

The bacteriologic study of otitis media revealed the isolation of a variety of organisms.
**Pseudomonas aeruginosa** is the most prominent organism being isolated in 40 (43.01%) of the cases. It is followed in order of predominance by Staphylococcus aureus in 31 (33.33%) cases. This is followed by E.coli is 12 (12.9%) cases. Proteus mirabilis in 5 cases (5.38%) Klebsiella pneumonia in 5 cases (5.38%).

Pseudomonas aeruginosa was the most predominant organism among the cases of CSOM reported by several workers in India and abroad with an incidence ranging from 21% to 52.94%.[10, 7,12] Staphylococcus aureus isolated in 31 cases (33.33%) was observed as the next predominant organism in this study. Our findings were similar to those of similar studies.[3,12 ]

Whereas, other studies had reported that Staphylococcus aureus was the most common etiology for CSOM. [5,13]

As regards the antibiotic sensitivity, in the present study Amoxiclav has proved to be the most effective drug for aerobes(59 strains) closely followed by Amikacin(55 strains), followed by Gentamycin(40 strains) and 38 strains sensitive to Erythromycin. Amongst the cases were Pseudomonas was isolated, maximum sensitivity was noted for Amikcin (37/40) followed by amoxiclav(25/40) and azithromycin (21/40). Least sensitivity was noted for magnex (13/40)

**CONCLUSION**

1. Ninety three patients with the clinical diagnosis of CSOM attending the out patient department of Narayana Hospital, attached to Narayana Medical College, Nellore formed the study group. They were investigated by direct smears and culture methods for the isolation of aerobic bacteria.

2. Monomicrobial aetiology was found to be the common among the study group.

3. Among the aerobes, *Pseudomonas aeruginosa* was the most common isolate (43.01%), the other organisms in the order of decreasing frequency were,

   - Staphylococcus aureus - 33.33%
   - Escherichia coli - 12.9%
   - Proteus vulgaris - 5.08%
   - Klebsiella - 5.08%

Monomicrobial infection was found to be common in this study.

The sensitivity pattern for *Pseudomonas aeruginosa* in decreasing order of frequency is Amikacin, Amoxiclav, Azithromycin, Gentamycin, Ciprofloxacin, Erythromycin, Neltimycin, and Magnex.

**REFERENCES**


