



Research Article

**STUDY ON MICROBIOLOGICAL PROFILE AND ANTIMICROBIAL SENSITIVITY PATTERN OF ISOLATED
PSEUDOMONAS AERUGINOSA IN CHRONIC SUPPURATIVE OTITIS MEDIA**

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ABSTRACT

Introduction: Chronic Suppurative Otitis Media (CSOM) is a potentially serious disease because of its complications. The incidence of CSOM is increasing in the developing countries because of the poor hygiene practices and lack of health education. *Pseudomonas aeruginosa* is a Gram-negative bacterium belonging to the family Pseudomonadaceae that is able to survive in a wide range of environments.

Aim: To determine the microbiological profile and antimicrobial sensitivity pattern of *Pseudomonas aeruginosa* from discharge in CSOM.

Material and method: This study was conducted over a time period of 6 months. All bacterial isolates were identified by conventional microbiological methods. Antimicrobial sensitivity testing of the isolates was done by Kirby-Bauer disc diffusion method according to Clinical and Laboratory Standards Institute (CLSI) guidelines.

Result: *Pseudomonas aeruginosa* (58.6 %) was the most prevalent bacteria isolated, followed by *Staphylococcus aureus* (19.57%). Most sensitive antibiotic against *Pseudomonas aeruginosa* was Piperacillin – Tazobactam (76.5%) followed by Meropenam (65.8%) and cephalosporins such as cephalexin, cefuroxime and amoxycillin-clavulanic acid shows complete resistance towards *Pseudomonas aeruginosa*.

Conclusion: It is important to evaluate bacteriologic profile and antimicrobial susceptibility pattern periodically for efficacious treatment in CSOM patients

KEYWORDS: CSOM, *Pseudomonas aeruginosa*, Antimicrobial Sensitivity, Aerobic bacterial culture.

INTRODUCTION

The world health organization (WHO) defines Chronic Suppurative Otitis Media (CSOM) as “a stage of ear disease in which there is chronic infection of middle ear cleft, a non - intact tympanic membrane (i.e. perforated ear drum) and discharge (otorrhea), for at least preceding 2 weeks”.

It is a disease of multiple etiologies and is well known for its persistence and recurrence in spite of treatment [1]. The

most common aerobic microorganism frequently isolated in CSOM is *Pseudomonas aeruginosa* and *Staphylococcus aureus*. In CSOM bacteria can reach middle ear from nasopharynx or external auditory canal through perforated tympanic membrane [2]. Among the various organisms isolated, *Pseudomonas aeruginosa* has been particularly blamed for deep seated and progressive destruction of middle ear and mastoid structures by releasing its toxins [3]. *Pseudomonas aeruginosa* is a Gram-negative bacterium belonging to the family Pseudomonadaceae that is able to survive in a wide range of environments [4]. The mechanism for antibiotics resistance can be classified into intrinsic, acquired and adaptive resistance. The intrinsic resistance of *P. aeruginosa* includes low outer membrane permeability, expression of efflux pumps that expel antibiotics out of the cell and the production of antibiotic inactivating enzymes. The acquired resistance of *P. aeruginosa* can be achieved by either horizontal transfer of resistance genes or mutational changes [5]. The adaptive resistance of *P. aeruginosa* involves formation of biofilm in the lungs of infected patients

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where the biofilm serves as a diffusion barrier to limit antibiotic access to the bacterial cells [6]. Development of complications depends on various reasons like high virulence of organism, poor resistance of patients, inadequate antibiotic treatment of acute middle ear and mastoid infection, presence of chronic systemic diseases and resistance of organisms to antibiotics which is becoming common on these days [7]. Untreated cases of CSOM results in complications such as persistent otorrhea, mastoiditis, labyrinthitis, facial nerve paralysis, intra cranial abscess, and thrombosis [8]. Empirical antibiotic therapy for *P. aeruginosa* includes either monotherapy or combination therapy, this therapy reduces the mortality in patients with severe *P. aeruginosa* infections [9]. However, treatment of *P. aeruginosa* infections has become a great challenge due to the ability of this bacterium to resist many of the currently available antibiotics [10].

It is important to know the major bacterial etiologies of CSOM and their antibiotic susceptibility patterns, both for selection of most appropriate treatment regimen and prevention of emergence of resistant strains [11].

MATERIALS AND METHODS

A prospective observational study was conducted in the department of ENT at Karuna Medical College Hospital, Vilayodi, Chittur, Kerala, over a period of 6 months from October 2018 to March 2019. This study was conducted after getting approval from the Institutional Human Ethical Committee (IHEC/09/2018) of KMCH. The patients attending in the ENT department with active purulent ear discharge and clinically diagnosed as Tubotympanic, Atticontral type CSOM and malignant otitis externa was included in this study. Patient has malignancy of middle ear treatment, previous history of surgery and Patient who already received treatment within 2 week for the same complaints of otitis media and malignant otitis externa were excluded from this study. Each of this patients are subjected to detailed clinical history regarding to name, age, sex, laterality and symptoms like ear discharge, hearing loss, vertigo, head ache, fever, nausea & vomiting, and facial weakness. Thorough ENT examination was done for each case by otoscopy and microscopic examinations. Pure tone audiometry (PTA) carried out for relevant cases. Two sterile cotton swabs were used to collect ear discharge from CSOM patients. One swab was used for performing gram stain and second one for culture. The culture was done on nutrient agar, blood agar, MacConkey agar and SDA. The organisms were identified by culture characteristics, morphology and conventional biochemical tests. Antimicrobial susceptibility

testing for *pseudomonas aeruginosa* was performed on Mueller-Hinton agar plate using Kirby-Bauer disc diffusion method. Collected data was entered in excel sheet for further evaluation and the results was presented in percentage. The main aim of the study was to determine the antimicrobial sensitivity pattern of *Pseudomonas aeruginosa* in CSOM patients.

RESULTS

In the present study of 'Bacteriological profile and drug sensitivity pattern of CSOM patients', conducted from October 2018 to March 2019; 123 patients fulfilling the inclusion criteria were randomly selected and analyzed.

Among the study population, 59 (47.9 %) were males and 64 (52.1 %) were females. The mean age of patients was 40.93 and the majority of patients (29.26 %) were aged 51-60 years. Most of the cases (40.65%) presented with right ear involvement.

A total of 246 swabs were collected from 123 cases and sent for gram staining and culture sensitivity. Culture of 123 swabs showed 25 (20.3%) were sterile while 98 swabs yielded growth of organisms. Out of 123 swabs, 3 (2.45%) yielded polymicrobial organisms, while 92 (74.80%) yielded mono microbial flora and three swabs shows fungal growth (2.45%) (Table 1).

To detect the presence and identify the general type of bacteria or sometimes fungi (microbes) in a sample taken from the site of a suspected infection; to generally classify bacteria grown in culture so that further identification tests can be performed and appropriate treatment given.

Polymicrobial isolation was associated with 6 organisms i.e. one combination of *Pseudomonas spp.* and *Klebsiella species*. Another two combinations include *Klebsiella spp.* with coagulase negative *staphylococcus aureus* and *streptococcus species*. So out of total microbes isolated (n=98), 73 were gram -ve bacteria and 25 were gram +ve organisms (Table 2).

Among the mono microbial isolates, *Pseudomonas aeruginosa* was the commonest offender seen in 54 (58.6%) swabs, followed by *Staphylococcus aureus* in 18 (19.57%) swabs. The most predominant organism causing CSOM among aerobic bacteria was *Pseudomonas aeruginosa*, followed by *Staphylococcus aureus* (Table 3).

Table No. 1: Culture pattern of ear discharge in CSOM patients.

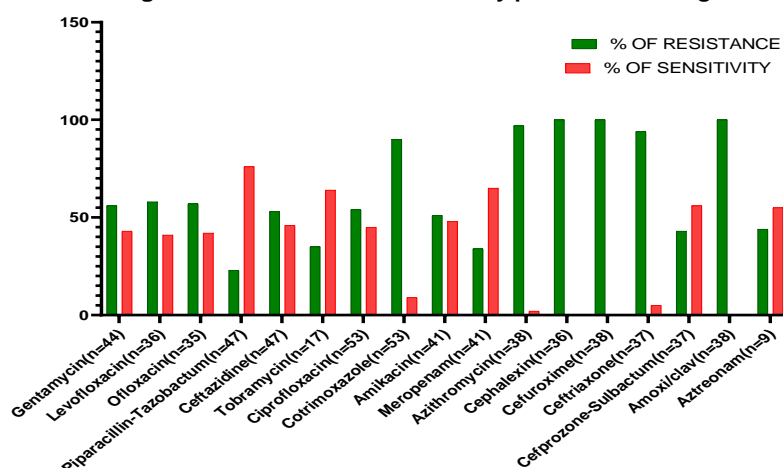
Pattern	Number (n=123)	Percentage (%)
No growth(sterile)	25	20.30
Fungal growth	3	2.45
Polymicrobial growth(mixed)	3	2.45
Monomicrobial growth (pure)	92	74.80

Table No. 2: Bacterial isolates according to gram staining.

Gram staining	Number of isolates (n=98)	Percentage (%)
Gram +ve	25	25.5
Gram -ve	73	74.4

Table No. 3: Bacteriological profile of CSOM cases (monomicrobial growth)

Organism	Number (n=92)	Percentage (%)
<i>Pseudomonas aeruginosa</i>	54	58.6
<i>Staphylococcus aureus</i>	18	19.57
CONS	9	9.78
<i>Klebsiella species</i>	2	2.17
<i>Streptococcus species</i>	4	4.35
<i>Acinetobacter species</i>	2	2.17
<i>Enterococcus species</i>	2	2.17
<i>Citrobacter species</i>	1	1.08

Fig.1: % of resistance and sensitivity pattern of *P. aeruginosa*Fig. 1: % of Resistance and Sensitivity Pattern of *P. Aeruginosa*Table No. 4: Antibiotic resistance and sensitivity pattern of *Pseudomonas Aeruginosa*

Antibiotics	No. of resistance	% of resistance	No. of sensitivity	% of sensitivity
Gentamycin (n=44)	25	56.81	19	43.18
Levofloxacin (n=36)	21	58.3	15	41.6
Ofloxacin (n=35)	20	57.14	15	42.85
Piparacillin-tazobactam (n=47)	11	23.5	36	76.5
Ceftazidime (n=47)	25	53.2	22	46.8
Tobramycin (n=17)	6	35.3	11	64.7
Ciprofloxacin (n=53)	29	54.8	24	45.2
Cotrimoxazole (n=53)	48	90.6	5	9.4
Amikacin (n=41)	21	51.3	20	48.7
Meropenam (n=41)	14	34.2	27	65.8
Azithromycin (n=38)	37	97.4	1	2.6
Cephalexin (n=36)	36	100	0	0
Cefuroxime (n=38)	38	100	0	0
Ceftriaxone (n=37)	35	94.5	2	5.5
Cefprozone-sulbactam (n=37)	16	43.3	21	56.7
Amoxi/clav (n=38)	38	100	0	0
Aztreonam (n=9)	4	44.5	5	55.5

Most of the *Pseudomonas* species were sensitive to piperacillin-tazobactam (76.5%). Meropenam were the second most effective group, showing 65.8% sensitivity followed by tobramycin (64.7%), and ceftazidime (56.7%) respectively. Cephalosporins such as cephalexin, cefuroxime and amoxycillin-clavulanic acid shows 100 % resistance to *P. aeruginosa*. Thus the most sensitive antibiotics to *P. aeruginosa* were piperacillin-tazobactam and the least effective

were cephalexin, cefuroxime and amoxycillin-clavulanic acid. (Table .4)

DISCUSSION

Chronic suppurative otitis media is one of the most common hearing problems, with approximately a 5% global incidence and is particularly prevalent in developing countries

[11]. Most common risk factors for CSOM are poor living conditions, poor access to medical care, inadequate medical treatment, recurrent upper respiratory tract infection and nasal disease [12].

In our study females are more commonly affected than males and which is in concordance with findings of other studies [13-15]. However, statistical analysis indicated that there were no significant differences between bacterial infections among the CSOM patients in terms of gender or age.

The highest no. of suspected CSOM patients were aged between 51-60 years and the highest bacterial infection were also found in this age group. Loy *et al.* showed that increased prevalence of csom patients in those aged 31-40 years [13]. In another study highest no. of CSOM cases were seen in 11-20 year age group [16].

Numerous studies show that majority of CSOM patients were aged less than 20 years [14, 17]. Analysis of bacteriology culture results from our study found pure culture (Monomicrobial) to be more common (74.80%) and this observation is supported by other investigators [17-20].

In the present study, out of total microbes isolated (n=98), 73 were gram -ve bacteria and 25 were gram +ve organisms. And which is in concordance with findings of other study which shows out of 90 culture smears, 59 (65.55%) were Gram negative and 31 (34.45%) were Gram positive [21].

In the present study, unilateral aural involvement was seen in most of the cases, of which, most of the cases (40.65%) presented with right ear involvement. Comparable results were obtained in the study of Gulati SP *et al* observed right side involvement in 51.5% cases.

Our study reveals that among the mono microbial isolates, *Pseudomonas aeruginosa* was the commonest offender seen in 54 (58.6%) swabs, followed by *Staphylococcus aureus* in 18 (19.57%) swabs. This result were comparable with study conducted by Bairy I *et al* which shows *Pseudomonas aeruginosa* (33.9%) as the commonest organism [22]. Similarly, *Pseudomonas* was also isolated as the predominant organism by Maji PK *et al* (63.8%) [23], Kumar S *et al* (45.9%) [24] and Indudharan R *et al* (27.2%) [25].

In our study *Citrobacter species* (1.08%) was found as the least isolates among monomicrobial organisms which is comparable with study conducted by Kumar R *et al* [26].

Pseudomonas aeruginosa showed higher resistance to cephalosporins such as cephalexin, cefuroxime and amoxycillin-clavulanic acid and it is supported by various studies like Ajay K *et al* [2] and higher sensitive to piperacillin-tazobactam (76.5%) followed by meropenam showing 65.8% sensitivity followed by tobramycin (64.7%), and ceftazidime-sulbactam (56.7%) respectively and it was comparable with study conducted by Mansoor T *et al* [27].

CONCLUSION

CSOM is one of the commonest chronic infectious diseases in developing countries. Factors playing role in the pathogenesis of CSOM are not fully comprehensible and underscore the urgent need for research in the area of development of novel and effective therapeutic strategies

Selection of antimicrobial agents must factor in the problem of drug resistance among infecting organisms. Hence, it becomes imperative on part of the health care facilities to undertake periodic evaluation of antimicrobial susceptibility profiles of the microbial pathogens for designing effective empiric treatment protocols and prevent potential risk of unforeseen complications.

We conclude that the *Pseudomonas aeruginosa* was moderately sensitive to Piperacillin-Tazobactam, Meropenam Tobramycin and Ceftazidime-sulbactam respectively. Resistance towards quinolones group of antibiotics increases when comparing with other studies. Regular antimicrobial sensitivity studies should be conducted as the microbial growth pattern changes with time because of indiscriminate and haphazard use of antibiotics. This study is more relevant in present scenario, because the knowledge of antimicrobial sensitivity pattern of microorganisms of CSOM will guide clinician in prescribing an empirical regimen so that a better and more specific management can be provided to CSOM patients.

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