



BACTERIOLOGY AND ANTIBIOTIC SUSCEPTIBILITY PATTERN OF CHRONIC SUPPURATIVE OTITIS MEDIA.

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ABSTRACT

Chronic suppurative otitis media (CSOM) is a chronic inflammation of the middle ear and mastoid cavity caused by bacteria and other microorganisms. It presents with hearing disability, and ear discharge through the tympanic membrane perforation. The disease is more common in developing countries due to poor socio-economic conditions, poor hygiene, poor nutrition, lack of education, and indiscriminate use of antibiotics, and lack of follow-up has led to the development of resistance strains. If left untreated leads to intra and extra-cranial complications. **Objective:-** The present study is undertaken to determine the causative bacteria and their antibiotic susceptibility pattern. **Material and methods:-** We studied 100 clinically suspected CSOM cases from January 2023 to August 2023 in the Microbiology and Otorhinolaryngology department of Basaweshwar Teaching and General Hospital Kalaburagi. Ear discharge was collected with a sterile swab under aseptic precaution from the external auditory canal (EAC) and the organism was grown on blood agar. The identification of organism and antibiotic susceptibility patterns was done by vitek2 compact automation. **Result:** A total of 100 cases were studied, among 66 were adults and 34 were children. The most common bacteria identified was pseudomonas aeruginosa 48(48%) followed by staphylococcus aureus (MRSA) 22(22%), other bacteria are staphylococcus haemolyticus 5(5%), Escherichia coli 5(5%), coagulase negative staphylococcus (CONS) 4(4%), klebsiella pneumoniae 2(2%) and others. Staphylococcus aureus was sensitive to linezolid, gentamicin, doxycycline, vancomycin, and pseudomonas aeruginosa was resistant to ciprofloxacin, tigecycline, and others. **Conclusion:-** The knowledge of microbial pattern and their antibiotic sensitivity helps in the rational use of antibiotics and prevents the emergence of resistant strains. It helps in accurate and quick diagnosis to guide appropriate treatment strategies.

KEYWORDS : Antibiotic drug resistance, CSOM, hearing disability, vitek2, MRSA.

INTRODUCTION

Chronic suppurative otitis media (CSOM) is defined as chronic inflammation of the middle ear and mastoid cavity that may present with recurrent ear discharge or otorrhea through the tympanic membrane perforation.

Risk factors include frequent episodes of acute otitis media, upper respiratory tract infections, and traumatic tympanic rupture as well as factors underlying the resource-limited living conditions such as overcrowding, malnutrition, poor hygiene, and chronic infectious diseases.²

Chronic suppurative otitis media (CSOM) is a common cause of hearing impairment, the worldwide prevalence of CSOM is 65-330 million people, and 39-200 million (60%) have clinically significant hearing impairment. The overall incidence is estimated to be around 9 per 100,000 people.³

CSOM is a disease with varied etiology and is classified into two types: mucosal type and squamous type of CSOM, depending on whether the disease process affects the pars tensa or pars flaccida of the tympanic membrane.³

In the literature, there are many causative agents for CSOM, but Pseudomonas aeruginosa and Staphylococcus aureus are the most common. The medical management of CSOM is usually empirical and based on topical antibiotics. Indiscriminate and inappropriate use of antibiotics to treat CSOM has led to the disease chronicity and the development of resistance strains.¹ Hence there is a need to treat the persistent ear discharge appropriately according to the antibiotic sensitivity pattern of the causative organisms.

Objective

1. The objective of the study was to identify the causative

bacterial strains that are responsible for the cases of CSOM.

2. To study their antibiotic susceptibility pattern mainly by automated method.



Figure : 1 Tympanic Membrane With Central Perforation.

MATERIAL AND METHODS

This is a prospective observational study conducted for 8 months (January 2023 to August 2023) in the Microbiology and Otorhinolaryngology (ENT) department of Basaweshwar Teaching and General Hospital Kalaburagi. The study was approved by our Institutional Ethical Committee and the written informed consent (signed by the patient or parent/guardian) was obtained at enrollment.

Inclusion Criteria

Patients with CSOM tubotympanic type with perforation who have not taken any treatment either systemic or topical ear drops for the last seven days.

Exclusion Criteria

Acute infection less than 3 months duration and CSOM with atticofacial type.

A total of 100 patients were recruited for the study. After cleaning the external ear canal, samples of middle ear discharge were aseptically collected from each patient by the ENT specialist using a sterile cotton swab and sent to the hospital's microbiology laboratory. Ear discharge samples were inoculated on Blood agar plates. Then, the plates were incubated at 37°C for 24 hours in aerobic conditions. Bacterial isolates were characterized by colony morphology, Gram staining, catalase, and coagulase test. Bacterial species were identified according to a standard microbiological procedure. The colony is picked and gram stain is done to identify gram-positive and gram-negative bacteria.

Pseudomonas Aeruginosa On Blood Agar And Gram Stain

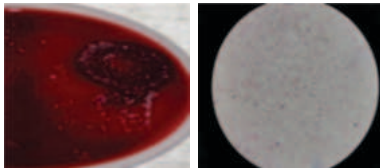


Figure:2 Klebsiella Pneumonia On Blood Agar And Gram Stain

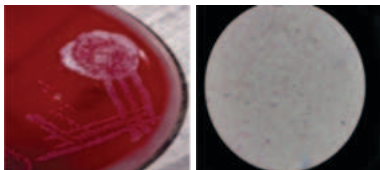


Figure:3 Staphylococcus Aureus On Blood Agar And Gram Stain

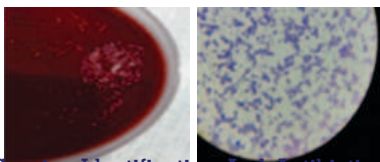


Figure:4 Species Identification And Antibiotic Sensitivity Testing Are Done By An Automated Method.

Vitek2 Procedure for Species Identification and AST

- 2 plastic tubes (no. 1 and no.2) were taken for isolation and placed on a cassette
- 3ml of suspension solution in each tube from the dispensator was taken
- tube1 as ID tube (for identification), and tube2 as AST tube (for sensitivity)
- picked similar isolated colonies from quadrant 4 from the plate, and made a suspension in tube1.
- fixed the turbidity of tube 1 up to 0.05-0.63 McFarland with the help of densi-chek plus.
- transferred 145µl for GNB or 280µl for GPS from tube1 to tube2
- placed ID card in tube1 and AST card in tube2
- Place the cassette in the vitek2compact machine and

readings are taken by using software.

- AST was done using vitek2 automation and MIC was recorded interpretation was done according to guidelines of clinical laboratory standard institute (CLSI), the AST panels for GPC P-628 and GNB N-280 and statistical analysis ST-03 were used.



Figure:5 vitek2 Machine

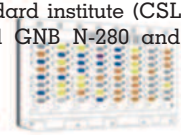
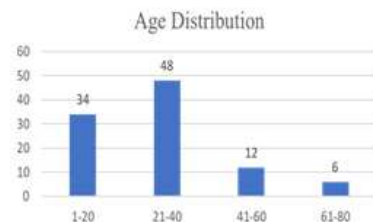


Figure:6 AST Card

RESULTS

The study sample (n=100) consists of the age group 1 to 80 years among these patients most common age group affected was the age group of 21 to 40 (48%), followed by 1 to 20 (34%), then 41-60 (12%), 61-80 (6%). Among 100 samples Gram-positive bacteria were observed in 37 (37%) and Gram-negative bacteria in 63 (63%) samples. The number of bacterial species identified according to their decreasing order of frequency are *Pseudomonas* spp. (48%), *Staphylococcus aureus* MRSA (22%), *Staphylococcus aureus* (6%), *Escherichia coli* (5%), *Staphylococcus haemolyticus* (5%), Coagulase-negative staphylococci CONS (4%), *Proteus mirabilis* (3%), *Proteus vulgaris* (3%), *Klebsiella pneumoniae* (2%), *Acinetobacter baumannii* (1%), *Enterobacter cloacae* complex (1%).



Graph1: Shows the Age Distribution of CSOM Patients

Antibiotic Susceptibility Testing

Table 1 shows the drug susceptibility pattern of gram-negative bacteria. The overall sensitivity rate was 79.1% and the highest sensitivity is seen with piperacillin/tazobactam combination 98.5%. The total resistance rate of gram-negative bacteria was 20.9% with resistance rates ranging from 1.5% to 73.1%. very high resistance was observed against tigecycline 73.1%. The antibiotics colistin, ciprofloxacin, and gentamicin showed resistance (42.8%, 38.1% and 34.9% respectively). very low resistance was detected with ceftazidime, amikacin, cefepime, Imipenem, cefoperazone/ sulbactam, piperacillin/tazobactam (12.7%, 9.6%, 3.2%, 3.2%, 3.2%, 1.5% respectively). The overall sensitivity rate was 79.1% and the highest sensitivity is seen with the piperacillin/tazobactam combination 98.5%.

Table-1 Drug Susceptibility Pattern of Gram-negative Bacterial Isolates

Drug	Pseudomonas aeruginosa (n=48)		Escherichia coli (n=5)		Proteus mirabilis (n=3)		Klebsiella pneumonia (n=2)		Proteus vulgaris (n=2)		ABC (n=2)		ECC (n=1)		Total n. (%) n=63	
	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R
TZP	48	0	4	1	3	0	2	0	2	0	2	0	1	0	62 (98.5)	1 (1.5)
SFP	48	0	3	2	3	0	2	0	2	0	2	0	1	0	61 (96.8)	2 (3.2)
IPM	48	0	3	2	3	0	2	0	2	0	2	0	1	0	61 (96.8)	2 (3.2)
MEM	48	0	3	2	3	0	2	0	2	0	2	0	1	0	61 (96.8)	2 (3.2)
AN	43	5	4	1	3	0	2	0	2	0	2	0	1	0	57 (90.4)	6 (9.6)
GM	38	10	3	2	3	0	2	0	0	2	2	0	1	0	41 (65.1)	22 (34.9)
CIP	30	18	2	3	3	0	2	0	0	2	2	0	1	0	39 (61.9)	24 (38.1)
TGC	08	40	4	1	0	3	2	0	0	2	2	0	1	0	17 (26.9)	46 (73.1)
CS	18	30	3	2	0	3	0	2	0	2	2	0	1	0	36 (57.2)	27 (42.8)

FEP	48	0	3	2	3	0	2	0	2	0	2	0	1	0	61(96.8)	2(3.2)
CAZ	48	0	2	3	3	0	0	2	0	2	2	0	0	1	55(87.3)	8(12.7)
Total	425	103	34	21	27	6	18	4	12	10	22	0	10	1	548	145
n. (%)	(80.5)	(19.5)	(61.8)	(38.1)	(81.8)	(18.2)	(81.8)	(18.2)	(54.5)	(45.5)	(100)	(0)	(90.9)	(9.1)	(79.1)	(20.9)

Table 2 Shows the drug susceptibility pattern of Gram-positive bacteria. The overall resistance rate of the Gram-positive bacteria was 39.1%, with the resistance rates ranging between 0% and 97.3%. Very high resistance was detected against Benzyl Penicillin G (97.3%). 70.3% of the Gram-positive bacteria were resistant to ciprofloxacin and levofloxacin. 50% or higher resistance rates were observed against amoxicillin/clavulanic acid, piperacillin/tazobactam, oxacillin, cefuroxime, cefotaxime, cefepime, imipenem, meropenem. Drugs like azithromycin, clarithromycin, Erythromycin, and clindamycin show resistance (62.1%, 62.1%, 62.1%, and 51.3% respectively). Very low resistance is shown by trimethoprim/sulfamethoxazole, nitrofurantoin, and rifampicin (27.1% 5.4%, and 2.7%, respectively). Overall sensitivity was 60.9% and 100% sensitivity are shown by vancomycin, linezolid, and daptomycin, teicoplanin, tetracycline, tigecycline.

TABLE-2 Drug Susceptibility Pattern of Gram-positive Bacterial Isolates

Drugs	Staphylococcus aureus (MRSA) N=22		Staphylococcus aureus N=6		Staphylococcus haemolyticus N=5		CONS N=4		Total (%) N=37	
	S	R	S	R	S	R	S	R	S	R
PG	0	22	1	5	0	5	0	4	1 (2.7)	36 (97.3)
AMC	0	22	6	0	5	0	4	0	15 (40.5)	22 (59.5)
TZP	0	22	6	0	5	0	4	0	15 (40.5)	22 (59.5)
OXI	0	22	6	0	5	0	2	2	13 (35.1)	24 (64.8)
CXM	0	22	6	0	5	0	4	0	15 (40.5)	22 (59.5)
CTX	0	22	6	0	5	0	4	0	15 (40.5)	22 (59.5)
FEP	0	22	6	0	5	0	4	0	15 (40.5)	22 (59.5)
IPM	0	22	6	0	5	0	4	0	15 (40.5)	22 (59.5)
MEM	0	22	6	0	5	0	4	0	15 (40.5)	22 (59.5)
GM	20	2	5	1	5	0	4	0	34 (91.8)	3 (8.1)
CIP	2	20	2	4	5	0	2	2	11 (29.7)	26 (70.3)
LEV	2	20	2	4	5	0	2	2	11 (29.7)	26 (70.3)
AZM	5	17	2	4	4	1	3	1	14 (37.8)	23 (62.1)
CTM	5	17	2	4	4	1	3	1	14 (37.8)	23 (62.1)
E	5	17	2	4	4	1	4	0	14 (37.8)	23 (62.1)
CM	8	14	3	3	5	0	2	2	18 (48.6)	19 (51.3)
LNZ	22	0	6	0	5	0	4	0	37 (100)	0
DAP	22	0	6	0	5	0	4	0	37 (100)	0
TEC	22	0	6	0	5	0	4	0	37 (100)	0
VA	22	0	6	0	5	0	4	0	37 (100)	0
TE	22	0	6	0	5	0	4	0	37 (100)	0
TGC	22	0	6	0	5	0	4	0	37 (100)	0
FT	22	0	6	0	5	0	2	2	35 (94.5)	2 (5.4)
RA	21	1	6	0	5	0	4	0	36 (97.3)	1 (2.7)
SXT	17	5	3	3	3	2	4	0	27 (72.9)	10 (27.1)
TOTAL	239 (43.5)	311 (56.5)	118 (78.6)	32 (21.3)	115 (92)	10 (8)	84 (84)	16 (16)	542 (60.9)	347 (39.1)

Abbreviations:- S-Sensitivity, R-Resistance, TZP-piperacillin/tazobactam, SFP-cefoperazone/ Sulbactam, IPM-imipenem MEM-meropenem AN-amikacin GM-gentamicin CIP-ciprofloxacin, TGC-tigecycline CS-colistin, FEP-cefepime, CAZ-ceftazidime, PG-benzyl penicillin, AMC-amoxicillin/clavulanic acid, OXI-oxacillin, CXM-cefuroxime, CTX-cefotaxime, LEV-levofloxacin, AZM-azithromycin, CTM-clarithromycin, ERY-erythromycin, CLL-clindamycin, LNZ-linezolid, DAP-daptomycin, TEC-teicoplanin VA-vancomycin, TEC-teicoplanin, TE-Tetracycline, FT-nitrofurantoin, TE-Tetracycline, RA-rifampicin, SXT-trimethoprim/sulfamethoxazole.

DISCUSSION

In the present study, all patients were suffering from a safe type of Chronic suppurative otitis media with central and subtotal perforation. The causes of CSOM were upper respiratory tract infection, unhygienic practices like ear pricking, swimming, trauma, inadequately treated ASOM, and others.

In this study, the maximum number of patients are in the age group of 20-40 (48%) followed by 10-20 (34%) years and patients were suffering from a moderate type of conductive hearing loss.

CSOM is one of the chronic infectious diseases worldwide,

commonly having its onset in childhood and causing considerable morbidity into adulthood. In CSOM, the causative bacteria *Pseudomonas aeruginosa* is known to cause deep-seated and progressive destruction of the middle ear and mastoid structures through its toxins and enzymes.

In our study, we observed the main causative organism was *Pseudomonas aeruginosa* followed by *staphylococcus aureus* (MRSA) strains and others.

Regarding the antibiotic susceptibility pattern, the organism *pseudomonas aeruginosa* showed almost 95-100% sensitivity to piperacillin/tazobactam, ceftazidime, cefoperazone/sulbactam, meropenem, amikacin, gentamicin and resistant to ciprofloxacin, levofloxacin, tigecycline, colistin.

The *staphylococcus aureus* (MRSA) almost showed 100% sensitivity to linezolid, daptomycin, teicoplanin, vancomycin, tetracycline, tigecycline, gentamycin and resistant to piperacillin/tazobactam, amoxicillin/clavulanic acid, meropenem, cefoperazone.

This is similar to the study conducted by Kombade, et al. The most common isolate found in this study was *Pseudomonas aeruginosa* followed by *Staphylococcus aureus*, and others.³ This study also provides insights into the susceptibility profile of bacteria isolated from ear infections. *Pseudomonas*

aeruginosa was found to be highly sensitive to ceftazidime, Colistin, piperacillin/tazobactam and ceftazidime.³

In contrast to this, in the study conducted by Rangaiah ST et al, *Staphylococcus* was the most common organism isolated. *Pseudomonas aeruginosa* was the second most common organism isolated so the causative organism varies according to demographic region. Antibiotic susceptibility testing was carried out for all the organisms that showed sensitivity to Meropenem, Piperacillin /Tazobactam, and Gentamycin. *Pseudomonas* isolates were most sensitive to Piperacillin/Tazobactam and *Staphylococcus aureus* isolates were most sensitive to Linezolid.

In a similar study conducted by Ismail Mohamed Ali et.al out of 200 samples showing bacterial growth, monomicrobial growth was detected in 176 (88%) and polymicrobial growth in 24 (12%) samples. Among 176 samples with monomicrobial growth, Gram-positive bacteria were observed in 40 (22.7%) and Gram-negative bacteria in 136 (77.3%) samples. The bacteriology of the samples with monomicrobial growth consisted of (in decreasing frequency) *Pseudomonas* spp. (50%), *Staphylococcus aureus* (15.9%), *Escherichia coli* (15.3%), Coagulase-negative staphylococci (6.8%), *Proteus mirabilis* (6.8%), and *Klebsiella* spp. (5.1%).

According to a report by WHO, India belongs to the highest (>4%) CSOM-prevalent countries. Hence, early diagnosis, knowledge of regional etiological agents, and an effective antibiotic policy can curtail the development of CSOM in fatal cases.³

CONCLUSION

In conclusion, *Pseudomonas aeruginosa* species and *Staphylococcus aureus* (MRSA) species were the most common isolates. *Pseudomonas* species showed the highest susceptibility to piperacillin/tazobactam, cefoperazone/sulbactam, imipenem, gentamicin, and *Staphylococcus aureus* showed the highest susceptibility to linezolid, daptomycin, vancomycin, tetracycline, tigecycline, gentamycin.

The most common associated risk factors were inadequately treated ASOM and poor hygiene like ear pricking and others. Culture and sensitivity are recommended before treating CSOM to evaluate the causative agent and its antibiotic susceptibility pattern for specific management.

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