ORIGINAL ARTICLE

Microbial Profile and Antibiogram of Bacteria Isolated from Chronic Suppurative Otitis Media in a Tertiary Care Hospital, Puducherry

Monica Edwin[1], Pramodhini S[1], Karthikeyan P[2], Umadevi S[1], Joshy M. Easow[1]*

[1]Department of Microbiology, [2]Department of ENT, Mahatma Gandhi Medical College and Research Institute, Sri Balaji Vidyapeeth (Deemed To Be University), Puducherry-607403 (Tamil Nadu), India

Abstract:

Background: Chronic Suppurative Otitis Media (CSOM) is a one of the major public-health problems and presenting with high prevalence rate in developing countries like India. Necessity to immediate intervention and treatment is required as the persistent infection is associated with increased risk of extracranial and intracranial complications. *Aim and Objectives:* This study was undertaken to determine the microbial profile of CSOM and also to detect the resistance pattern of bacterial pathogen isolated. *Material and Methods:* This prospective analytical single centre study was done on patients presenting with ear discharge of more than 6 weeks duration. All the samples were processed for aerobic bacterial and fungal culture and antibiotic susceptibility testing was done by Kirby Bauer Disc Diffusion Method. *Results:* Analysis of 85 pus samples from CSOM showed 97.6% were culture positive and 2.3% were culture negative. Of 83 culture positive samples, 79 (95.2%) were mono-microbial and 4 (4.8%) were polymicrobial. The predominant organisms isolated from CSOM was *Pseudomonas spp* (52.9%), followed by *Staphylococcus aureus* (20.7%). Antimicrobial sensitivity testing showed that 10.7% of Gram negative bacilli were Extended Spectrum Beta-Lactamase (ESBL) producers and 16.6% of *Staphylococcus aureus* was Methicillin resistant and 33.7% of the 86 isolates were resistant to Ciprofloxacin. *Conclusion:* In conclusion, our study on microbial profile of CSOM showed *Pseudomonas aeruginosa* was predominantly isolated followed by *Staphylococcus aureus* and around 45% of the isolates were resistant to Ciprofloxacin. Knowledge on the predominant isolates and their antibiotic susceptibility pattern of the bacteria isolated from CSOM is very crucial for starting the empirical treatment, prior to availability of susceptibility reports from the laboratory.

Keywords: Chronic Suppurative Otitis Media, *Pseudomonas aeruginosa, Escherichia coli*, Extended Spectrum Beta-Lactamase

Introduction:

Chronic Suppurative Otitis Media (CSOM) is an inflammation of the middle ear and mastoid mucosa with perforation of tympanic membrane. It is further classified based on disease effects into tubotympanic and atticoantral type [1]. Presence of continuous discharge along with tymphanic perforation for periods from 6 week to 3 months is recognized as CSOM cases.The aerobic bacteria commonly isolated from CSOM are *Pseudomonas aeruginosa, Staphylococcus aureus* followed by *Proteus mirabilis, Klebsiella pneumoniae, Escherichia coli* etc., and the anaerobic bacteria isolated includes *Bacteroides spp, Peptostreptococcus spp, Peptococcus spp* etc [2]. Infection of the middle ear and external auditory canal by fungi is common as most of the fungi have predilection for moist pus. *Candida species* and *Aspergillus species* are the most commonly isolated fungal species. Based on geographical and various other

factors, there occurs variation in the type of organism isolated [3]. This study has been undertaken to determine the microbial profile of chronic suppurative otitis media and also to detect the resistance pattern of bacterial pathogen isolated.

Material and Methods:

This prospective analytical single centre study was carried out for a period of six months from 1st September 2019 to 28th February 2020 as a short-term project in a tertiary care hospital. Sample size was calculated based on past one-year record from our Microbiology laboratory data. This project was carried out after approval from Institutional Ethics Committee. Pus samples were collected from patients presenting to ENT Department with tympanic perforation and ear discharge of more than 6 weeks duration in a tertiary care hospital after informed consent from the patient. Ear discharge associated with tympanic membrane perforation collected from patient for a period of more than 6 weeks duration was included in this study. Those presenting with tympanic membrane perforation and ear discharge of less than 6 weeks duration months were excluded from this study.

Two sterile cotton swabs of ear discharge from each patient were collected aseptically and sent to Microbiology laboratory for processing. The samples were then inoculated into blood agar and MacConkey Agar, incubated at 37°C. For fungal culture, samples were inoculated in Sabouraud's Dextrose Agar (SDA) and incubated at 25°C. Identification of the bacterial isolates was done as per standard protocol, such as colony morphology, Gram staining and the biochemical reactions [4]. Bacterial susceptibility to antimicrobial agents was carried out by Kirby Bauer Disc Diffusion Method as per Clinical and Laboratory Standards Institute (CLSI) Guidelines [5]. Identification of growth in the SDA tubes was done based on colony morphology and microscopic examination with Lactophenol Cotton Blue (LPCB) mount according to standard procedures [6]. In case of yeast, germ tube formation test done to identify *Candida albicans* and *non albicans Candida.*

Statistical Analysis:

Data were entered in MS-Excel 2010, percentages were calculated for categorical variables and analysed.

Results:

In our study, out of total 85 cases, 50.6% were males and 49.4% were females and 49% was maximum number had fallen between the age group of 18-30 years (Table 1). Analysis of 85 pus samples from CSOM showed 97.6% were culture positive and 2.3% were culture negative. Out of 83 culture positive samples, 79 (95.2%) were monomicrobial and 4 (4.8%) were polymicrobial as shown in Table 2. A total of 87 microbes isolated from 83 culture positive samples, of which 75.9% were Gram negative, 22.9% were Gram positive bacteria and 1.1% were *Candida*. The predominant organisms isolated from CSOM were *Pseudomonas spp* (52.9%), followed by *Staphylococcus aureus* (20.7%), *Escherichia coli* (8%), *Klebsiella pneumonia* (4.6%), Non-fermenting Gram negative *bacilli* (4.6%), *Proteus spp* (3.4%), *Citrobacter spp* (2.3%), *Enterococci* (2.3%) and *Candida albicans* (1.1%) (Table 3).

The sensitivity of *Staphylococcus aureus* was maximum to clindamycin (100%), tetracycline (94%) and Cotrimoxazole (89%) and maximum resistance was to ciprofloxacin (66.7%).

Antimicrobial sensitivity testing showed that 10.7% of Gram negative bacilli were Extended Spectrum Beta-Lactamase (ESBL) producers and 16.6% of *Staphylococcus aureus* was methicillin resistant (Tables 4 and 5) *Pseudomonas aeruginosa* showed 100% sensitivity to Imipenem and Meropenem, 98% to piperacillin and 93% to amikacin and 67% resistance to Ciprofloxacin (Table 6). Overall, 33.7% of the 86 isolates were resistant to Ciprofloxacin.

Table 1: Age-sex Distribution of the CSOM (n=85)

Age	Male	Female	Number (%)
18-30	18 (42.9%)	24 (57.1%)	42 (49.0%)
31-45	13 (86.7%)	2 (13.3%)	15 (18.0%)
46-60	5 (41.7%)	7 (58.3%)	12 (14.0%)
61-75	7 (43.8%)	9 (56.2%)	16 (19.0%)
Total	**43(50.6%)**	**42(49.4%)**	**85 (100.0%)**

Table 2: Pattern of Isolation of Microbial Growth and Analysis of Isolates from CSOM

Analysis of isolates	Number	Percentage
Total number of patients	**85**	**100**
Culture positive	83	97.6
Culture negative	2	2.3
Total number of isolates	**87**	100
Gram negative bacteria	66	75.9
Gram positive bacteria	20	22.9
Fungus	1	1.1
Monomicrobial	79	95.2
Polymicrobial	4	4.8

Table 3: Microbial Flora of CSOM

Isolates	Number	Percentage
Gram negative bacilli		
Pseudomonaspp	46	52.9
Escherichia coli	7	8.0
Klebsiella pneumoniae	4	4.6
Citrobacterspp	2	2.3
Proteusspp	3	3.4
Non-fermenting Gram negative bacilli	4	4.6
Gram Positive cocci		
Staphylococcus aureus	18	20.7
Enterococci	2	2.3
Fungus		
Candida albicans	1	1.1
Total	87	100.0

Table 4: Antimicrobial Susceptibility Pattern of Gram-Positive Organisms in CSOM (n=20)

Antibiotics	*Staphlococcus aureus* (n=18)				*Enterococci* (n=2)			
	S	%	R	%	S	%	R	%
Penicillin	2	11.1	16	88.9	1	50	1	50
Cotrimoxazole	16	88.9	2	11.1	1	50	1	50
Erythromycin	10	55.6	6	33.3	0	0	2	100
Clindamycin	18	100.0	0	0.0	-	-	-	-
Tetracycline	17	94.4	1	5.6	0	0	2	100
Oxacillin	15	83.3	3	16.7	-	-	-	-
Gentamicin	12	66.7	6	33.3	2	100	0	0
Ciprofloxacin	6	33.3	12	66.7	1	50	1	50

Table 5: Percentage of Antimicrobial Sensitivity Pattern of Enterobacteriaceae in CSOM (n=16)

Antibiotics	*Escherichia coli* (n=7)		*Klebsiella pneumoniae* (n=4)		*Citrobacter spp* (n=2)		*Proteus spp* (n=3)	
	S%	R%	S%	R%	S%	R%	S%	R%
Ampicillin	57	43	-	-	50	50	100	0
Cotrimoxazole	100	0	75	25	100	0	67	33
Ceftriaxone	71	29	75	25	100	0	67	33
Amikacin	86	14	100	0	100	0	100	0
Gentamicin	100	0	75	25	100	0	67	33
Tobramycin	0	0	0	0	0	0	0	0
Ciprofloxacin	29	71	100	0	50	50	33	67
Imipenem	100	0	100	0	100	0	100	0
Meropenem	100	0	100	0	100	0	100	0
Piperacillin Tazobactum	100	0	100	0	100	0	100	0

Table 6: Percentage of Antimicrobial Sensitivity Pattern of Non Fermenting Gram Negative Bacilli in CSOM (n=50)

Antibiotics	*Pseudomonas spp* (n=46)		Non fermenting gram negative bacilli (n=4)	
	S%	R%	S%	R%
Ceftazidime	93	7	75	25
Amikacin	93	7	75	25
Gentamicin	87	13	75	25
Tobramycin	85	15	100	0
Ciprofloxacin	67	33	50	50
Imipenem	100	0	100	0
Meropenem	100	0	100	0
Piperacillin Tazobactum	98	2	100	0

Discussion:

CSOM is a one of the major public-health problems and presenting with high prevalence rate in developing countries like India. Immediate intervention and treatment are required as the persistent infection is associated with increased risk of extracranial and intracranial complications. Infection can spread from middle-ear to vital structures such as mastoid, facial nerve, labyrinth, lateral sinus, meninges and brain leading to both intracranial and extracranial complications [7]. The disease incidence is maximum among developing countries in particular among people belonging to low socio-economic status because of underlying risk factors like undernutrition, overcrowding, inadequate health care, poor personal hygiene and recurrent episodes of upper respiratory tract infection [8].

In our study, out of total 85 cases, males (50.6%) were more affected than females (49.4%). Our data was in consonance with the study by Ahmed *et al.* [9], were they showed male (57.29%) predominance than females (42.70%). It was observed in the present study, that prevalence was high among the age group of 18-30 years (49%). Study by Jyothi *et al.* [10], observed 50% of cases in the age group of less than 20 years, which was concordant with our study. Studies have shown predominance of age group affected were less than 10 years with 43% [11] and 22.93% [2].

In the present study, 97.6% were culture positive and only 2.3% were sterile. This result of this study correlated to the study conducted by Jain *et al*. [2], where they had 96.18% specimens were culture positive and 3.82% were culture negative. Few other similar studies showed 12 (6.25%) and 13 (5.28%) culture negative samples. Monomicrobial growth was observed in 95.2% of the cases, which correlates to the study by Agarwal *et al*. [12]. Polymicrobial growth was reported in 4.8% of the samples studied whereas study by Jangla *et al.* [13], observed 11% polymicrobial growth.

In the present study, the predominant organisms isolated from CSOM was *Pseudomonas spp* (52.9%), followed by *Staphylococcus aureus* (20.7%). Microbial profile of CSOM by Jain *et al*. [2], reported among bacteria, *Pseudomonas aeruginosa* was most common (42.17%) followed by *Staphylococcus aureus* (29.93%) well concordant with our findings. Various other similar studies, have reported predominant growth of *Pseudomonas aeruginosa* followed by *Staphylococcus aureus* [10, 14, 15].

As we observed the bacterial profile of CSOM was similar and concurrent with several studies, but there occurred changing trends of antimicrobial susceptibility pattern of isolated organisms over a period of time. The sensitivity of *Staphylococcus aureus* was maximum to clindamycin (100%), tetracycline (94%) and cotrimoxazole (89%) and maximum resistance was to ciprofloxacin (66.7%). *Pseudomonas aeruginosa* showed 100% sensitivity to imipenem and meropenem, 98% to piperacillin and 93% to amikacin and 67% resistance to Ciprofloxain, which is comparable to study by Hiremath *et al.* [16] where they observed maximum resistance of 78.9% and Bagwan *et al*. [17] observed 77% resistance to ciprofloxacin. Antimicrobial susceptibility pattern of showed that 10.7% of Gram-negative *bacilli* were ESBL producers and 16.6% of *Staphylococcus aureus*

were methicillin resistant and 44.1% of the 86 isolates were resistant to Ciprofloxacin. In a similar study by Yogeesha *et al.* [18], *Pseudomonas spp* had shown 33% resistance to ciprofloxacin and overall Gram negative bacilli showed 44.1% resistance to ciprofloxacin. Since ciprofloxacin is the commonly and inadvertently used topical antibiotics, there is now increasing trends of resistance.

Conclusion:

In conclusion, our study on microbial profile of CSOM showed *Pseudomonas aeruginosa* was predominantly isolated followed by *Staphylococcus aureus* and around 45% of the isolates were resistant to ciprofloxacin. The knowledge on the most common organisms associated with the CSOM and their antibiotic sensitivity pattern, plays an important role in formulating an antibiotic policy and to start empirical therapy prior to the availability of culture and sensitivity report. Alarming rising trends of multi-drug resistant organisms, necessitates the need for continuous strict surveillance of resistant bacteria and in turn to avoid the indiscriminate use of antibiotics.

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Author for Correspondence:
Dr. Pramodhini S, Department of Microbiology, Mahatma Gandhi Medical College and Research Institute, Sri Balaji Vidyapeeth (Deemed To Be University) Pondicherry 607402
Email: pramodhinis@gmail.com Cell: 9445216145

How to cite this article:
Edwin M, Pramodhini S, Karthikeyan P, Umadevi S, Easow JM. Microbial Profile and Antibiogram of Bacteria Isolated from Chronic Suppurative Otitis Media in a Tertiary Care Hospital, Puducherry. *J Krishna Inst Med Sci Univ* 2020; 9(4):72-79

Submitted: *16-Aug-2020* ***Accepted:*** *25-Sept-2020* ***Published:*** *01-Oct-2020*