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Original Research Article

Aerobic Bacterial and Fungal Profile of Chronic Suppurative Otitis Media and its Antibiotic Susceptibility Patter

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Abstract:

Background and Objectives: Chronic suppurative otitis media is defined as chronic inflammation of middle ear andmastoid cavity that presents with recurrent ear discharge of more than three months duration through a perforated tympanic membrane. CSOM is a major health problem in developing countries causing serious local damage and threatening complications. To isolate and to identify aerobic bacteria from Chronic suppurative Otitis media cases, to study antibiotic susceptibility pattern of isolated aerobes and facultative anaerobes, to isolate and identify fungi causing CSOM and to study the antifungal susceptibility pattern of isolated *Candida* species.

Materials and Methods: This study was conducted at the Department of Microbiology, at DMCH, Darbhanga. A total of 500 patients with clinical diagnosis of CSOM from the Department of ENT were enrolled in the study and the samples were obtained from each patient using sterile cotton swabs processed by standard microbiological techniques.

Conclusion: Chronic suppurative otitis media is a major health problem in our setup causing serious local damage and threatening complications. Pseudomonas aeruginosa followed by Staphylococcus aureus are the most common bacterial isolates causing CSOM.

Keywords: *Staphylococcus aureus,* Pseudomonas aeruginosa*,* CSOM.

Introduction

Chronic suppurative otitis media is defined as chronic inflammation of middle ear andmastoid cavity that presents with recurrent ear discharge of more than three months duration through a perforated tympanic membrane.[1] CSOM is a major health problem in developing countries causing serious local damage and threatening complications. It is an important cause of preventable hearingloss in developing countries. The infection may begin in childhood as a complication of untreated or inadequately treated ASOM or may be chronic in onset. The bacteria may gain entry to the middle ear through a chronic perforation.[2] The spread of micro-organisms to the adjacent structures of the ear may cause local damage or intracranial and extra cranial complications, ranging from persistent otorrhoea, mastoiditis, labyrinthitis, facial nerve palsy, meningitis, intracranial abscess or thrombosis and sepsis.[3] Children tend to have a higher predisposition to ear infections than adults because the anatomy of the Eustachian tube which is shorter, more horizontal with more flaccid cartilage and permits easier access of organisms through the nasopharynx.[2] Incidence of this disease is higher among people with low socioeconomic status because of malnutrition, overcrowding, poor hygiene, inadequate health care, and recurrent upper respiratory tract infection. It is a massive health problem in developing countries and India is one of the countries with highest CSOM prevalence (>4%) where urgent attention is needed.[1] According to WHO survey, about 65-330 million people suffer from ear infection worldwide and 60% of them have had significant hearing impairment.[3] It causes conductive and sensorineural hearing loss and has got adverse effect on childhood development and may have long term effects on early communication, language development, auditory processing, educationalprocess and physiological and cognitive development.[4] It has profound impact on society in terms of resources utilised in treatment and the direct impact the chronic infection has on hearing of patients.[5] CSOM is classically divided into 1) a tubotympanic type affecting the middle ear muco-periosteum and 2) an attico-antral type which is an active squamous disease with a growth of squamous epithelium into the middle ear cleft.6 Recently CSOM has been classified into following types; active mucosal, inactive mucosal, active squamous, inactive squamous and healed (dimeric, tympanosclerosis etc).[7] The most common microorganisms found in CSOM are

Pseudomonas aeruginosa, Staphylococcus aureus, Klebsiella pneumoniae, Proteus mirabilis, Escherichia coli, Aspergillus spp., Candida spp. and these vary in different geographical distributions. Fungal infections of the middle ear are common as fungi thrive well in moist pus. [8] Therefore, this study was undertaken to know the pattern of microbes and their antimicrobial susceptibility pattern to provide a guideline for empirical antibiotic therapy. Though the treatment of CSOM is controversial, and subject to change particularly in developing countries, the antibiogram of these organisms have been reported to vary with time and geographical area, probably due to use and abuse of antibiotics. Hence the need for periodic update of antibiogram for effective chemotherapy and management of CSOM cannot be overemphasized.

Objectives

To study antibiotic susceptibility pattern of isolated aerobes and facultative anaerobes. To isolate and identify fungi causing CSOM.

To study the antifungal susceptibility pattern of isolated Candida species.

Materials and Methods

This prospective study was conducted in the Department of Microbiology at darbhanga Medical College and Hospital Darbhanga Bihar.

Study duration of Eighteen Months. Department of Otorhinolaryngology, DMCH Darbhanga. 500 patients with clinical diagnosis of CSOM attending OPD and IPD of ENT department of DMCH. who satisfied the inclusion criteria were enrolled for the study.

All ear discharge samples from cases of CSOM with a minimum of 500 consecutive non-duplicate isolates will be included. Sample size has been calculated to be 500 depending upon previous years statistics in our institution.

Table 1:

Number of cases/day	1-2
Number of cases/month	25-30
Number of cases/18 months	450-500

Inclusion Criteria

- Patients diagnosed as CSOM after thorough clinical evaluation i.e. the patients having ear discharge of more than 3 months.
- Patients who were not on antibiotic (both systemic and topical) treatment for minimum of 24 hours prior to sample collection.

Exclusion Criteria

- Patients suffering from CSOM who are on systemic antibiotics in the past 7 days of presentation.
- Patients who are on topical medications to the ear.

Patients having ear discharge due to some traumatic or neoplastic condition

The swab on reaching the laboratory was inoculated on the following culture media

- 5% Sheep Blood agar plate
- MacConkey agar plate
- Thioglycollate broth

After overnight incubation at 37^0C aerobically, the plates were examined for growth and culture characteristics were identified. The isolates were identified by Grams stainmorphology, motility, culture characteristics and biochemical reactions by the standard techniques. The isolated colonies depending on the Gram reaction were subjected to following biochemical tests for identification.

If gram positive cocci catalase test was done, for catalase positive tube coagulase was done -Tube coagulase positive were Staphylococcus aureus whereas tube coagulase negative were Coagulase negative staphylococcus aureus(CONS).

Pseudomonas aeruginosa

Gram stain

Gram negative bacilli

Culture

Table 2: Culture

Blood Agar	MacConkey Agar
Large grey, translucent colonies withspreading periphery and beta haemolysis with fruity odour.	Non lactose fermenting colonies withspreading periphery, pigmentation if present was noted.

Preliminary tests

Table 3: Preliminary tests

Oxidase test	Positive
Catalase test	Positive
Hanging drop	Motile
OF Test	Oxidative
Growth at 44 degrees	Positive

Biochemical reactions

Table 4: Biochemical reactions

Mannitol motility agar	Mannitol not fermented, motile
Triple sugar iron agar	Alkaline slant/Alkaline butt
Indole test	Negative
Nitrate reduction test	Positive
Citrate test	Positive
Urease test	Negative
Arginine	dihydrolysed

Escherichia co

Gram stain

Gram negative straight rods

Culture

Table 5: Culture

Blood Agar	MacConkey Agar
Grey translucent, convex colonies	Large nonmucoid lactose fermenting colonies

Preliminary tests

Table 6: Preliminary tests

Oxidase test	Negative
Catalase test	Positive
Hanging drop	Motile
OF Test	Fermentative

Results

Total 500 patients were included in the study. 63% of the patients were males while 37% of patients were females. Highest incidence of CSOM was found to be among patients in the age group of 20-29 years (32.8%) while the lowest incidence was found in patients ≥ 60 years age (3.2%).53% of total male patients were in age group of <10yrsand 20-29yrs .400 patients (80%) were outpatients while 100 patients (20%) were inpatients.

Table 7: Age and Sex wise distribution of patients (n=500)

Age (Yrs.)	Males	Females	Total	Percentage
<10	58	26	84	17%
10-19	53	30	83	16.5%
20-29	108	56	164	32.8%
30-39	42	33	75	15%
40-49	38	20	58	11.8%
50-59	16	4	20	4%
≥60	9	7	16	3.2%
Total	315	185	500	100%

The most common presenting complaint was ear discharge for more than 3 months (93%), followed by decreased hearing (66%). Upper respiratory tract infection was noted in 30% cases and allergic history was noted in 12% cases. 25% cases had a past history of modified radical mastoidectomy with tympanoplasty(Type 2). Tonsillitis and DNS was noted in 5% and 3% respectively. Tuberculosis and adenoids werenoted in 2% cases. Stroke(1%), Miller-Fischer syndrome(1.6%), Extrapulmonary tuberculosis(0.4%) and nasal carcinoma (0.4%) were the other co-morbidities among the patients.

Table 8: Diseases associated with CSOM among patients

Associated features	Number of cases	Percentage(%)
Urti	150	30%
Allergy	64	12%
Past History of Modified RadicalMastoidectomy	130	26%
Tonsillitis	26	5%
Dns	14	3%
Tb	10	2%
Adenoids	10	2%

The most common organism isolated was *Pseudomonas aeruginosa* (44%) followed by *Staphylococcus aureus* (22%). Methicillin resistant *staphylococcus aureus* was isolated in 15 cases (3%). CONS were isolated in 40 cases (8%). Fungi were isolated in 6% cases with *Aspergillus niger* isolated in 13 cases (43.4%), *Aspergillus flavus* isolated in 5 (16.7%), *Aspergillus fumigatus* isolated in 2 cases (6.7%), *Candida albicans* isolated in 4(13.3%), *Candida parapsilosis* in 4(13.3%), *Candida tropicalis* in 1(3.3%) and *Candida lusitaniae* isolated in 1 case(3.3%). In mixed culture growth, the most common isolates were *Pseudomonas aeruginosa* and *Klebsiella pneumoniae* (2.8%) followed by *Pseudomonas aeruginosa* and *E.coli* (2.6%).

Table 9: Culture results in patients (n=500)

Type of organism	Total cases	Total isolates	Percentage
Pure growth	350	350	70%
Mixed growth	75	150(75×2)	15%
No growth	30	0	6%
Commensals	45	45	9%
Total	500	545	100%
Candida Sp.	10		2%
Aspergillus Sp.	20		4%
Total	500		100 %

Table 10: Distribution of bacterial and fungal isolates (n=500)

	Number	Percentage
Bacterial isolates	470	94%
Fungal isolates	30	6%
Total	500	100%

Table 11: Distribution of Gram positive bacteria(n=153)

Gram Positive Bacteria	No. Of Isolates	Percentage
Staphylococcus aureus (MRSA + MSSA)	113	73.85%
CONS	40	26.15%
Total	153	100%

MDR was defined as acquired nonsusceptibility to at least one agent in three or more antimicrobial categories.

In present study, a Gram-negative bacterium was considered MDR when it is resistant to representative drug from these three groups of antibiotics, β-lactam (ceftazidime), aminoglycoside (gentamicin) and quinolone (ciprofloxacin). Among the Carbapenemase (+) Pseudomonas aeruginosa isolates,2(14.3%) were MDR (+) whereas 12(85.7%) were MDR(_).Among Non-carbapenemases,8(3.4%) were MDR(+) whereas 198(96.6%) were non-MDR. P value 0.1263 which is not statistically significant.

Discussion

Chronic Suppurative Otitis Media(CSOM) is considered as a major public health problem in the developing world and India is one of the countries with high prevalence where urgent attention is needed.[1] It is a persistent disease with risk of irreversible complications and is an important cause of preventable hearing loss in adults and children.[9] Since chronic suppurative otitis media is a disease which can cause significant morbidity, early microbiological diagnosis ensures effective treatment.[10] Hence knowledge of pathogens and their antibiotic susceptibility pattern would guide the treating physicians in selection of appropriate antibiotics which would help us in reducing the complications and emergence of resistant strains.[11] In the present study, 545 isolates were obtained from 500 patients. CSOM was found to be more common among males than females with males accounting for 63% cases. This may be a reflection of the overall male predominance of childhood infections due to anatomic, behavioural and socioeconomic differences between males and females.[2] The most common presenting complaint in our study was ear discharge for more than 3 months (93% cases), followed by decreased hearing (66%). Upper respiratory tract infection was noted in 30% cases and allergic history was noted in 12% cases. 26% cases had a past history of modified radical mastoidectomy with tympanoplasty. In the present study, fungi were isolated among 30 patients. Among the fungal isolates, *Aspergillus species* accounted for 66.7% isolates and *Candida* species accounted for 33.3% isolates. Among *Aspergillus Sp*., *Aspergillus niger* was the most common isolate (43.4%) followed by *Aspergillus flavus* (16.7%) and *Aspergillus fumigatus* (6.7%). *Candida albicans* was isolated in 4 cases(13.3%), *Candida parapsilosis* in 4cases(13.3%), *Candida tropicalis* in 1 case (3.3%) and *Candidalusitaniae* in 1 case (3.3%). Similar study was conducted by Sen Gupta et al who reported 74.2% of *Aspergillus species* and 19.3% of *Candida species* among the fungal isolates.[15] The second most commonly isolated organism in our study was *Staphylococcusaureus* comprising of 113(22%) of the isolate. 96 isolates (85%) of *Staphylococcus aureus* were found to be methicillin sensitive(MSSA) whereas 17 isolates(15%) were found to be MRSA. 100% isolates of *Staphylococcus aureus* were sensitive to Linezolid and

Teichoplanin. 65% of *Staphylococcus aureus* isolates were resistant to Ciprofloxacin and 54% were resistant to Amoxicillin- Clavulanate. This indicates high level of resistance among *Staphylococcus aureus* for β-Lactams and Fluoroquinolones . In a study done by Prakash et al out of 181 isolates, 48% of isolates were *Staphylococcus aureus* and all were foundto be methicillin sensitive.[12] CSOM is generally treated by oral medications with quinolone antibiotic drops such as Ciprofloxacin. The absence of oral formulations of Piperacillin, Ceftazidime and Imipenem has severely limited the use of these antibiotics in patients with CSOM, thus restricting the ability of these bacteria to develop resistance to these antibiotics. This may explain why resistance to Imipenem in bacteria from CSOM otorrhea is lower than in other types of bacterial infection.[13,14] The present study has showed that CSOM infections are commonly caused by *Pseudomonas aeruginosa* followed by *Staphylococcus aureus*. However *Pseudomonas* are becoming less sensitive against commonly used antimicrobials like Ciprofloxacin, Cephalosporins, and Gentamycin. Resistance to antimicrobial agents isan increasing public health threat. It limits therapeutic options and leads to increased morbidity and mortality. The important factor that is responsible for resistance is inappropriate use of antibiotics. It is essential to follow antibiotic policies which guides judicious and appropriate use antibiotics to prevent emergence and spread of resistant pathogens. Hence continuous and periodic evaluation of microbiological pattern and antimicrobial sensitivity of the isolates is necessary to decrease the risk of complications and emergence of resistant microorganisms by early institution ofappropriate treatment.

Conclusion

The results of this study indicate that ESBL and Amp C production is a majormechanism of resistance to cephalosporins among the isolates of Gram-negative bacteria. The emergence of organisms possessing combinations of β-lactamases like ESBL and Amp C beta lactamases, Carbapenemases is a major public health concern necessitating efficient detection and intervention to control drug resistance. This study also shows that bio-film production may play an important role in recurrence and antibiotic resistance in unsafe CSOM. This study also shows that fungi also account for a small proportion of cases ofCSOM in our setup. Identification of fungi and antifungal susceptibility is important for appropriate treatment of such cases. Hence continuous and periodic evaluation of microbiological pattern and antimicrobial sensitivity is essential to reduce the potential risk of complications and emergence of resistant strains.

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