Original Article

Microbiology of Chronic Suppurative Otitis Media in a Tertiary Care Setup of Uttarakhand State, India

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Abstract

Background: Chronic suppurative otitis media (CSOM) is a notorious infection and a major health problem in developing countries causing serious local damage and threatening complications. Early and effective treatment based on the knowledge of causative micro-organisms and their antimicrobial sensitivity ensures prompt clinical recovery and possible complications can thus be avoided. **Aims:** The aim of this study was to isolate the organisms associated with CSOM and to detect the antibiogram of the aerobic isolates. **Materials and Methods:** A total of 204 patients clinically diagnosed of CSOM were enrolled in the study and the samples were obtained from each patient using sterile cotton swabs and cultured for microbial flora. Drug susceptibility testing for aerobic isolates was conducted using Kirby-Bauer disc diffusion method. **Results:** The most common causative organisms isolated were *Staphylococcus aureus* (48.69%) and *Pseudomonas aeruginosa* (19.89%) amongst the 191 aerobic isolates. Anaerobes accounted for 29.41% of the isolates while 12.25% were fungi. Antimicrobial profile of aerobic isolates revealed maximum sensitivity to amikacin (95.5%), ceftriaxone (83.4%) and gentamicin (82.7%). **Conclusion:** Knowing the etiological agents of CSOM and their antimicrobial susceptibility is of essential importance for an efficient treatment, prevention of both complications and development of antibiotic resistance and finally, the reduction of the treatment costs.

Keywords: Attico-antral, Chronic suppurative otitis media, *Pseudomonas aeruginosa, Staphylococcus aureus,* Tubotympanic

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Introduction

Chronic suppurative otitis media (CSOM) is defined as chronic inflammation of middle ear and mastoid cavity that may present with recurrent ear discharges or otorrhoea through a tympanic perforation.[1] Incidence of this disease is higher in developing countries especially among low socio-economic society because of malnutrition, overcrowding, poor hygiene, inadequate health care, and recurrent upper respiratory tract infection.[2] The urban to rural ratio of the disease is 1:2 and the poorer rural communities have highest prevalence.[2,3]

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CSOM is usually classified into two types, tubotympanic and attico-antral depending on whether the disease process affects the pars tensa or pars flaccida of the tympanic membrane (TM).[2] Tubotympanic is called as a safe type or benign type as there is no serious complication whereas, attico-antral is called as the unsafe or dangerous type because of associated complication and may be life threatening at times.[4] Infection can spread from middle-ear to vital structures such as mastoid, facial nerve, labyrinth, lateral sinus, meninges and brain leading to mastoid abscess, facial nerve, paralysis, deafness, lateral sinus thrombosis, meningitis and intracranial abscess.[5,6] Of all the complications, hearing loss associated with chronic ear discharge is nearly always significant, reported in 50% of cases and tending to be more severe than those reported in other types of otitis media.[7] Complications associated with CSOM were frequent in pre-antibiotic era, however, the introduction of antibiotics gave clinicians a tool to be used even without the precise etiological diagnosis and the irrational use of antibiotics led to the emergence

of multi-drug resistant bacterial strains and disease complication in return.[8] Changes in bacterial flora in CSOM in the last decade have been confirmed and described by various authors.[2,8,9]

The treatment of CSOM is controversial and subject to change particularly in the developing countries, the prevalence and antibiogram of these organisms has been reported to vary with time and geographical area as well as continent to continent, probably due to indiscriminate use of the antibiotics.[8] Hence, the periodic update of prevalence and antibiogram of the etiological agents for CSOM would be helpful in therapy and management of patients.

The objective of this cross sectional prospective study was to determine the microbial diversity and the resistogram of aerobic bacterial isolates among the patients suffering from CSOM who attended ENT Department of our hospital, a tertiary care center located in foothills of Himalayas. To the best of our knowledge no such data is available from this part of India.

Materials and Methods

This community based, prospective study was conducted for a period of 6 months (January 2012 to June 2012) in a Tertiary Care Hospital of Uttarakhand State, India. The study was approved by our Institutional Ethical Committee and the written informed consent (signed by patient or parent/guardian) was obtained at enrollment.

A total of 204 patients clinically diagnosed of CSOM, who did not received antimicrobial therapy (topical or systemic) for the last 7 days were included in the study. Ear discharge was obtained from the diseased ear of the patient, using three separate pre-sterilized swabs. One of the swabs was used for aerobic culture and was plated on 5% sheep blood agar (BA), MacConkey's agar and chocolate agar (CA). The plates were incubated at 37°C for 48 h.

Second swab used for anaerobic culture was inoculated in Robertson's cooked meat (RCM) broth and incubated at 37°C for 72 h. On 3rd day, sub-cultures from RCM were made on 5% BA and Neomycin BA (Neomycin at a working concentration of 70 µg/ml). A metronidazole disc (5 µg) was placed at the junction of secondary and tertiary streaking area, opposite to primary well of inoculation. Dynox anaerobic jar (Dynamicro Pvt. Labs Ltd., Mumbai, India) based on Marshal's chromous absorption principle[10] was used for anaerobic culture. The jars were closed and incubated at 37°C for 72 h and thereafter, examined for the zone of inhibition around the metronidazole disk. An aertolerance test on CA was set up to rule out facultative anaerobes.

Third swab was used for mycological culture and was inoculated on two slants of Sabouraud Dextrose Agar with chloramphenicol (0.05%) and were then incubated at 28°C and 37°C. The slants were later examined for gross and the microscopic morphology of the fungi.

Organisms were identified using standard procedures.[11,12] Antimicrobial sensitivity testing for aerobic isolates was carried out by Kirby Bauer disc diffusion method on Muller Hinton agar. Results were interpreted in accordance with central laboratory standards institute guidelines.[13]

All dehydrated media, reagents and antibiotic discs were procured from Hi-media Laboratories Pvt. Ltd., Mumbai, India.

Statistical analysis

The data was analyzed by using Statistical Package for Social Sciences (SPSS) version 11 and the prevalence of organisms was determined and expressed in percentage.

Results

Out of total 204 ear swabs processed microbial growth was seen in 186 (91.18%) while 18 (8.82%) samples showed no growth. In 118 (57.84%) samples mono-microbial growth was seen whereas 68 (33.33%) samples showed poly-microbial growth. The mean age of the patients was 25.6 and the peak incidence of CSOM was observed in age group between 0 year and 20 years (51%) Table 1 shows the age wise distribution of various morphotypes in CSOM. Females (53.92%) were more commonly affected than males (46.08%) and the sex ratio female:male was 1.2:1.

Aerobic flora was seen in 174 (85.29%) samples in which 191 aerobic isolates were grown, 60 (29.41%) cases showed anaerobic flora and 69 anaerobes were recovered from them, 25 (12.25%) cases had fungal etiology from

Table 1: Age wise distribution of various morphotypes in chronic suppurative otitis media

Age	Morphotypes			Total
	Monomicrobial	Polymicrobial	Sterile	
0-10	28	20	2	50
11-20	24	24	6	54
21-30	22	10	2	34
31-40	18	2	4	24
41-50	10	6	0	16
51-60	8	0	2	10
61-70	0	4	2	6
71-80	8	2	0	10
Total	118	68	18	204

which 48 fungal isolates were obtained. Table 2 depicts the characterization of aerobic, anaerobic, and fungal isolates among CSOM patients.

Antimicrobial sensitivity testing was carried out for 181 isolates as 10 isolates were identified as Diptheroids. Results of sensitivity testing are depicted in Table 3. Amikacin (AK) (95.5%), ceftriaxone (CAX) (83.4%) and gentamicin (GEN) (82.7%) showed maximum activity to most of the isolates.

Table 2: Distribution of various isolates in chronic suppurative otitis media

Type of isolate	Frequency (%)
Aerobic isolates	
MSSA	93 (48.69)
Pseudomonas aeruginosa	38 (19.89)
Klebsiella pneumoniae	18 (9.42)
Escherichia coli	14 (7.33)
Diptheroides	10 (5.23)
Acinetobacter baumanii	6 (3.14)
CONS	4 (2.10)
Proteus vulgaris	4 (2.10)
Pneumococci	2 (1.05)
Morganella morgagnii	2 (1.05)
Total	191 (100)
Anaerobic isolates	
Clostridium species	18 (26.09)
Peptococcus species	16 (23.19)
Peptostreptococcus species	16 (23.19)
Prevotella melaninogenica	11 (15.94)
Bacteroides species	8 (11.59)
Total	69 (100)
Fungal isolate	
Aspergillus niger	18 (37.50)
Aspergillus fumigatus	16 (33.33)
Candida albicans	6 (12.50)
Candida species	08 (16.67)
Total	48 (100)

MSSA: Methicillin sensitive *Staphylococcus aureus*; CONS: Coagulase negative *Staphylococcus* species

Discussion

CSOM is a major public-health problem, and India is one of the countries with high-prevalence where urgent attention is needed.[1] It's a persistent disease with great risk of irreversible complications. CSOM is an important cause of preventable hearing loss particularly in the developing world[5] and a reason of serious concern, particularly in children, because it may have long-term effects on early communication, language development, auditory processing, educational process, and physiological and cognitive development.[1] Early, microbiological diagnosis ensures prompt and effective treatment to avoid such complications. High prevalence of culture positive cases of CSOM (91.18%) was seen in the present study. We found that the CSOM was more prevalent in first and second decade of life and accounted for 51% of the cases. This finding corroborates well with the observations made by other researchers.[14-18] High-prevalence of CSOM in children may be attributed to the fact that they are more prone to upper respiratory tract infections (URTIs). Furthermore, cold weather pre-disposes children to URTI.[19,20] Poor hygiene and unorthodox approach to treatment like use of unconventional ear drops and concoctions such as oil and honey into the middle-ear may initiate the proliferation of opportunistic pathogens leading to blockage of eustachian tube (ET).[21]

The male is to female ratio was found to be 1.2:1. Cases of CSOM were more common in females (110) than in males (94). This observation was parallel with the findings of few authors[18,22] and in contrast with other researchers.[2,16] As this study involved, a random selection of cases the predominance of female patients over male may be only an incidental finding. Moreover, no knowledge of anatomical differences in the ear structures of male and female has been reported.

Analysis of the total 204 cases revealed that mono-microbial growth was obtained in 118 (57.84%)

Table 3: Sensitivity pattern of aerobic bacterial isolates in chronic suppurative otitis media

Isolate tested	No. of isolate	AMP	AC	GEN	AK	CZ	CXM	CAX	CTX	CIP
MSSA	93	79	87	83	89	87	87	89	81	77
Pseudomonas aeruginosa	38	27	NT	31	36	NT	NT	22	28	30
Klebsiella pneumoniae	18	Nil	10	11	16	06	06	14	10	16
Escherichia coli	14	06	08	09	14	06	08	12	12	10
Acinetobacter baumanii	06	02	04	05	06	04	04	06	04	06
Proteus vulgaris	04	02	02	04	04	02	02	02	02	02
CONS	04	04	04	03	04	04	04	04	04	02
Pneumococci	02	02	02	NT	NT	02	02	02	02	02
Morganella morgagnii	02	Nil	Nil	02	02	Nil	Nil	Nil	02	Nil
Total	181	122	117	148	171	111	113	151	145	145

MSSA: Methicillin sensitive *Staphylococcus aureus*; CONS: Coagulase negative *Staphylococcus* species; AMP: Ampicillin; AC: Amoxycillin clavulanic acid; AK: Amikacin; CZ: Cefazolin; CXM: Cefuroxime; CAX: Ceftriaxone; CTX: Cotrimoxazole; CIP: Ciprofloxacin; GEN: Gentamicin

samples, 68 (33.33%) samples yielded polymicrobial growth, whereas, 18 (8.82%) samples showed no growth. Corresponding figures reported by other authors vary significantly. Aslam, *et al*. from Pakistan[23] in their study on 142 samples revealed that 76% of them were pure and 23.9% were mixed cultures and only 2.1% fungi, whereas, Poorey and lyer from India[16] in their study on 100 samples found pure growth from 82, mixed growth from 10, and no growth in 8 samples. Difference in results of various authors could have been due to the difference in the patient population studied and geographical variations. In the present study, mono-microbial etiology was found to be more common (51.84%) and this observation was supported by other researchers.[16-18,22] A study from Iran[24] reported mono-microbial etiology to be 100% in all 61 samples studied. In contrast, some researchers found polymicrobial etiology more prominent in otitis media.[21,25]

Predominant bacterial etiology (aerobic) of CSOM in this region is *Staphylococcus aureus* (48.69%) and this observation was in line with diversity of microbial flora of CSOM infection in colder regions as reported in studies by Ettehad, *et al*.[24] from Iran (31.15%) and Singh, *et al*.[26] from India (36%). In contrast, other studies from India,[2,17] Nigeria,[14] and Pakistan[18] showed different trends as Pseudomonas was the most prevalent organism and this could be due to the variation in micro-organisms in different regions and effect of climate. In our study, we could isolate Pseudomonas in 19.89% of cases. Pseudomonas, however, is the predominant cause of CSOM in tropical region does not usually inhabit the upper respiratory tract, its presence in the middle-ear cannot be ascribed to an invasion through ET and it should be considered as secondary invader gaining access to the middle-ear via defect in TM.[8,27]

Coliforms including *Klebsiella pneumoniae* and *Escherichia coli* were isolated from 9.42% and 7.33% cases respectively, and these findings were tandem to the reports by Mansoor, *et al*.[18] who reported the same to be 8% and 4% whereas Poorey and lyer.[16] reported a high-incidence for klebsiella in their study (25.4%). A recent study by Shyamala and Reddy from India[17] showed a little different trend where *E. coli* was reported in 12% and Klebsiella in 5% of cases. More frequent isolation of fecal bacteria like *E. coli*, Klebsiella and water bacteria like Pseudomonas indicates that individuals are at high-risk of infection due to poor hygiene conditions. Our study revealed that both gram-positive and gram-negative organisms are responsible for infection of middle-ear. It is usually seen that gram-negative rods outnumber the gram-positive organisms in CSOM as reported by various authors.[2,17,18] However, in our study *S. aureus* was the predominant organism followed by Pseudomonas and other gram-negative rods and this observation corroborates well with reports by other researchers.[26,28]

The role of anaerobes in CSOM is often questioned as they are mostly detected in cases with extensive cholesteatoma or granulation tissue however, it is advocated that while investigating pathogenic organisms in CSOM requests for anaerobic culture should be included and the medical therapy should be directed at the eradication of the pathogenic aerobic and anaerobic organisms.[29,30] Out of total 204 samples anaerobic etiology was found in 60 (29.41%) samples and 69 anaerobic isolates were yielded. Our findings were in contrast to Ibekwe, *et al*., Maji, *et al*. and Indudharan, *et al*.[29,31,32] who found negligible anaerobic isolates in their studies. However, a study by Srivastava, *et al*.[33] from India found anaerobic etiology in 10.2% of cases. Another study from Iran[34] reported, the same to be 14.3%. Various anaerobic organisms isolated are shown in Table 2. The most predominant were Clostridium species (18 isolates), Peptococcus species and Peptostreptococcus species (16 isolates each). Growth of anaerobes showed that progression of infection from acute to chronic stage could have created anaerobiosis, which offered favorable microenvironment for the growth and proliferation of anaerobic pathogens. There was strong evidence that mixed aerobic, anaerobic cultures characterized chronic infection suggesting a potential synergy between anaerobic and aerobic bacteria. It has been previously reported that polymicrobial infections are more pathogenic than mono-microbial infections.[35]

Fungal infections of the middle-ear are common as fungi thrive well in moist pus. The most commonly found fungi in CSOM are Candida species and Aspergillus species.[29] In the present study, fungal etiology was found in 25 (12.25%) cases out of which 29.17% were Candida species (*Candida albicans*-6, Non-albicans Candida-8) and 70.83% were Aspergillus species (*Aspergillus niger*-18, *Aspergillus fumigatus*-16). In a study from Haryana, India,[2] fungal etiology was found in 15% of cases, out of which 60% were Candida species and 40% were Aspergillus species. In another study from Singapore[14] on 90 patients of otitis media, fungi accounted for 8.8% of the total isolates out of which Aspergillus species was found in 33.3% followed by Candida species 22.2%. These findings may be attributed to the environmental effects on the cases of otitis media, which were studied in this area.

Antimicrobial susceptibility test (AST) was carried out for all the aerobic isolates (except for 10 isolates of Diptheroids). AK was found to be most effective drug followed by CAX, GEN and ciprofloxacin (CIP). These findings were parallel to the reports by other authors.[2,18,36] For the antibiotics commonly available as topical ear drops, GEN, and CIP showed good activity for

most of the commonly isolated organism and can be used as effective first line topical antibiotic in the treatment of CSOM. Studies have revealed that quinolones like CIP are safe and effective particularly against *S. aureus* and *Pseudomonas aeruginosa*.[37-39]

Isolation of various aerobic, anaerobic, and fungal isolates shows that different conditions of CSOM could be differentiated on microbiological grounds. Thus, for better management of CSOM, microbial classification of infection as well as drug sensitivity test of organism recovered are essential for making appropriate decision of antimicrobials that will effectively eradicate the pathogen.

When results of our study were compared with the findings of other researches, it was clear that microbial profile and AST pattern of CSOM has been changing with due course of time. Geographical variation and difference in-patient population studied could be the possible factor for variability. Emergence of antimicrobial resistance is becoming more common. Indiscriminate and haphazard antibiotic use as well as negligence on patient part are the factors responsible. As the symptoms subside many patients stop taking antibiotics before the completion of therapy and allow the partly resistant microbes to flourish. Patients should be instructed to avoid such practice. Changes in the microbial flora following the advent of sophisticated synthetic antibiotics increase the relevance of and reappraisal of the modern day flora in CSOM and there *in vitro* AST pattern is very important for the clinician to plan the treatment of a chronically discharging ear.

Conclusion

CSOM like other chronic disease can limit an individual's employability and quality of life. Experts declare that when prevalence of CSOM is > 3% it must be targeted as a high-priority disease.[1] In many developing countries, including India, prioritizing health-care need is a difficult task. As long as health-care delivery fails to target high-risk groups in developing countries, as of ours infections like CSOM will persist. With the development and widespread use of antibiotics, the types of pathogenic micro-organisms and their resistance to antibiotics have changed. Continuous and periodic evaluation of microbiological pattern and antibiotic sensitivity of isolates is necessary to decrease the potential risk of complications by early institution of appropriate treatment. As higher incidence of disease was seen among children so educating parents and guardians on possible risk-factors of the disease may be a preventive strategy that might reduce disease occurrences. We believe that our data may contribute to an effective management of CSOM.

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