Aerobic Bacterial and fungal profile with antimicrobial susceptibility in patients of CSOM attending tertiary care hospital

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Abstract

Chronic Suppurative Otitis Media (CSOM) is a long standing infection of a part or whole of the middle ear cleft characterized by ear discharge & various complications if not treated especially in developing countries with low socio-economic society because of malnutrition, overcrowding, poor hygiene and inadequate health care & laboratory facilities. Study was conducted to identify the common isolates from patients diagnosed with CSOM at tertiary centre in central India and to determine the antibiotic sensitivity pattern of these bacterial isolates to commonly used antimicrobials.

A total of 157 patients were enrolled after IEC approval and processed for isolation of bacterial and fungal isolates using standard microbiological tests with sensitivity pattern. Data was compiles and statistical analysis was performed by using Statistical Package for the Social Sciences‖ (SPSS) software - 16 and MS Microsoft Excel 2007. A total of 151(96.18%) samples were culture positive. Bacterial cultures were obtained in 84.71% of the cases whereas polymicrobial growth (aerobic bacteria + fungi) were seen in 11.47% cases. The bacterial isolates were mostly sensitive to commonly used antimicrobials like 3rd generation cephalosporins, Amoxycillin+Clavulanic acid, Amikacin & Ciprofloxacin and multi drug resistant organisms were not observed. More than 90 % of the yeasts were sensitive to commonly used antifungal agents.

Evidence based judicial treatment & appropriate use of antimicrobials in an environment where antibiotics are commonly abused will ensure & will reduce the resistance to antimicrobials and the cost of treatment.

Keyword: Chronic Suppurative otitis media, Ear discharge, Antibiotic sensitivity.

Introduction

Chronic Suppurative Otitis Media (CSOM) is a long standing infection of a part or whole of the middle ear cleft characterized by ear discharge and a permanent perforation. 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.(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)

Prevalence surveys, estimate that the global burden of illness from CSOM may involve 65 to 330 million individuals with draining ears. According to the 2004 WHO report on CSOM highest prevalence (more than 6%) was found in Greenland, India, the United Republic of Tanzania and the Solomon Islands, and among the Australian aborigines.(3)

Most common isolates found in CSOM are Aerobic bacteria (*Pseudomonas aeruginosa, Staphylococcus aureus, Proteus mirabilis, Klebsiella pneumonia, Escherichia coli* etc.), anaerobic bacteria (*Bacteroids, Peptostreptococcus, Peptococcus* etc.*)* and fungi *Aspergillus* species, *Candida* species etc. but distribution of these isolates vary in various geographical areas.(4)

The basic principle of medical management of CSOM are aural hygiene and the use of topical antimicrobial agents but the indiscriminate and haphazard use of antibiotic and poor follow-up of these patients has resulted in emergence of multiple resistant strains of bacteria and the persistence of low grade infections.(5) The changes in the microbiological flora following the haphazard use of antibiotics have increased the relevance of the reappraisal of the flora in CSOM(6) and so it is very important for a clinician to know the antibiotic sensitivity pattern to plan for treatment.

Recognizing the need for awareness of the current pattern of bacterial pathogens isolated from CSOM in our setting, we conducted a study to identify the common isolates from patients diagnosed with chronic suppurative otitis media at PCMS & RC (People's Medical College Of Medical Sciences & Research Centre) Bhopal (M.P) and to determine the antibiotic sensitivity pattern of these bacterial isolates to commonly used antimicrobials.

Materials and Method

After approval of the Institutional Ethics Committee around 157 patients suffering from Chronic Suppurative Otitis Media (CSOM) attending the ENT department of the tertiary care hospital were enrolled with detailed clinical history for the cross sectional analytical study conducted during November 2012 to

April 2014. Ear discharge from clinically diagnosed enrolled patients was collected after written informed consent and were processed immediately in Microbiology Department.

The discharge was collected using three sterile cotton swab stick, labelled and taken to the laboratory immediately for processing.

The 1st swab was used to make a smear on a clean glass slide for direct smear examination by Gram's stain, Ziehl-Neelsen stain, and KOH mount.

2nd Swab was used to inoculate MacConkey's agar (MA) & Blood agar plates. All plates are incubated aerobically at 37^0C and evaluated at 24 hours, 48 hours and 72 hours and the plates are discarded if there is no growth. The specific identification of bacterial pathogens is done based on microscopic morphology, staining characteristics, cultural and biochemical properties using standard laboratory procedures.[7,8,9]

3rd swab was processed for the isolation of fungus and was cultured on Sabouraud Dextrose Agar (SDA) tubes in duplicate. One tube was incubated at room temperature 25^0C and another tube was incubated at 37^0C and observed daily for fungal growth upto 2 weeks.

Microscopic examination like LCB mount and slide culture were done to identify the fungi. Gram staining was done for identification of yeast and yeast like cells. Chlamydospore formation and germ tube tests were done to identify *Candida albicans.*

Antimicrobial susceptibility of the isolates was performed on Mueller Hinton Agar plates by Kirby-Bauer disc diffusion method according to the Clinical Laboratory Standards Institute (CLSI) guidelines.[10] Data was compiles and statistical analysis was performed by using Statistical Package for the Social Sciences (SPSS) software - 16 and MS Microsoft Excel 2007.

Result

Of the 157 cases studied cases 92(58.60%) cases were males and 65(41.40%) cases were females.84 were males and 66 females. Age of the subjects in the study group varies from 1 year to 73 years. Maximum patients of CSOM belongs to age group 11-20 years which was 37(23.57%) and least cases of CSOM which was13(8.28%) found in age group 41-50 years. All of the culture was negative in 0-10 years of age group. **[Table 1]**

Table 1: Age wise distribution of study population

Age	No. of patients	Percentages (%)
0-10 years	36	22.93
11-20 years	37	23.57
21-30 years	27	17.19
31-40 years	23	14.65
41-50 years	13	8.28
> 50 years	21	13.38
Total	157	100.00

Out of 157(100.00%) samples, 151(96.18%) samples were culture positive and 6 (3.82%) samples were culture negative. Out of 6(100.00%) culture negative samples 4(66.67%) samples obtained from male patients and 2(33.33%) from female patients. Here, a positive case referred to a case in which either the bacterial or fungal isolate was obtained.

The results of the mycological and the bacteriological studies on the 157 cases **[Table 2]** showed that positive bacterial cultures were obtained in 84.71% of the cases and polymicrobial growth (aerobic bacteria + fungi) were seen in 11.47% cases. Fungal isolates alone were not present in any of the cases whereas 3.82% cases were negative for both fungi and bacteria.

Table 2: Distribution of isolates among study population

Type of isolates	Frequency	Percentage(%)
Bacterial isolates	133	84.71
Bacterial and fungal isolates	18	11.47
No organism	6	3.82
Total	157	100.00

Out of 147 bacterial isolates *Pseudomonas aeruginosa* was most common 62(42.17%) followed by *Staphylococcus aureus* 44(29.93%). Among 18 fungal isolates Aspergillus niger8(44.44%) was most common followed by Cancida albicans 7(38.89%) and Candida non albicans 3(16.67%) **[Table 3].**

Table 3: Distribution of various Bacterial isolates in study population

Organism		Frequency(%)
Bacterial Isolates	*Pseudomonas aeruginosa*	62(42.17)
	Staphylococcus aureus	44(29.93)
	Klebsiella species	11(7.48)
	Escherichia coli	10(6.80)
	Streptococcus Pyogenes	06(4.08)
	Citrobacter freundii	04(2.72)
	Proteus mirabilis	04(2.72)
	Proteus vulgaris	04(2.72)
	Acinetobacter species	02(1.36)
	Total	147(100)
Fungal Isolates	*Aspergillus niger*	08(44.44)
	Candida albicans	07(38.89)
	Candida non albicans	03(16.67)
	Total	18(100)

Table 4 & 5 shows the antibiotic sensitivity pattern of various bacterias isolated from CSOM cases. Out of 147(100.00%) isolates 127(86.39%) were sensitive to Cefotaxime, 125(85.03%) were sensitive to Amoxyclav, 110(74.83%) were sensitive to Amikacin, 70(47.62%) were sensitive to Ciprofloxacin, 64(43.54%) were sensitive to Gentamicin and 59(40.14%) were sensitive to Cotrimoxazole. that Cefotaxime had highest sensitivity 43.30% for *Pseudomonas aeruginosa* followed by Amoxyclav 52(41.60%) and Amikacin 46(41.82%).

Table 4: Antibiotic sensitivity pattern for bacterial isolates (n=147)

Antibiotic	Sensitive N(%)	Total N(%)
Amikacin	110(74.83)	147(100.00)
Ciprofloxacin	70(47.62)	147(100.00)
Amoxyclav	125(85.03)	147(100.00)
Gentamicin	64(43.54)	147(100.00)
Cefotaxime	127(86.39)	147(100.00)
Cotrimoxazole	59(40.14)	147(100.00)

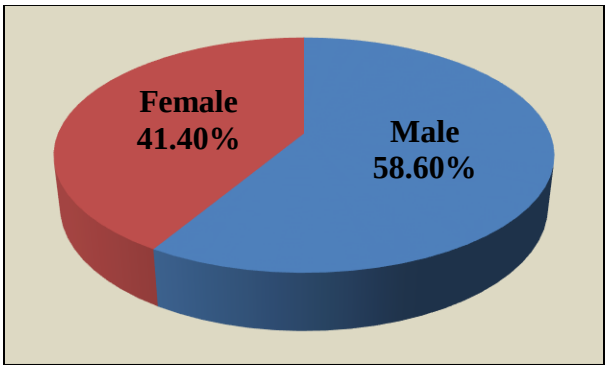

Fig. 1: Sex wise Distribution of study population

Table 5: Sensitivity pattern of bacterial isolates in CSOM

Isolates	No. of isolates N (%)	Type of Antibiotics N (%)					
		AK	CIP	CTX	GEN	AMC	COT
Pseudomonas aeruginosa	62 (42.17)	46 (41.82)	34 (48.58)	55 (43.30)	23 (35.94)	52 (41.60)	21 (35.59)
Staphylococcus aureus	44 (29.93)	34 (30.90)	23 (32.85)	39 (30.70)	19 (29.69)	42 (33.60)	21 (35.59)
Klebsiella species	11 (7.48)	8 (7.27)	4 (5.71)	7 (5.51)	9 (14.06)	9 (7.20)	5 (8.48)
Escherichia coli	10 (6.80)	8 (7.27)	3 (4.29)	10 (7.87)	7 (10.94)	8 (6.40)	8 (13.56)
Streptococcus Pyogenes	6 (4.08)	2 (1.82)	2 (2.86)	4 (3.51)	0 (0.00)	4 (3.20)	0 (0.00)
Citrobacter freundii	4 (2.72)	4 (3.64)	4 (5.71)	4 (3.15)	4 (6.23)	4 (3.20)	2 (3.39)
Proteus mirabilis	4 (2.72)	4 (3.64)	0 (0.00)	2 (1.57)	2 (3.12)	4 (3.20)	2 (3.39)
Proteus vulgaris	4 (2.72)	4 (3.64)	0 (0.00)	4 (3.15)	0 (0.00)	0 (0.00)	0 (0.00)
Acinetobacter species	2 (1.36)	0 (0.00)	0 (0.00)	2 (1.57)	0 (0.00)	2 (1.60)	0 (0.00)
Total	147 (100.00)	110 (74.82)	70 (47.62)	127 (86.93)	64 (43.54)	125 (85.03)	59 (40.14)

AK-Amikacin, CIP-Ciprofloxacin, CTX-Cefotaxime, GEN-Gentamicin, AMC-Amoxyclav, COT-Cotrimoxazole

The overall antifungal susceptibility for the *candida* isolated was highest 10 (100%) for Amphotericin-B, followed by Fluconazole, Itraconazole, Ketoconazole 9 (90%) each, Clotrimazole 8(80%) & Nystatin 7 (70%). **[Table 6]**

Table 6: Antifungal sensitivity pattern for fungal*(Candida)* isolates (n=10)

Antifungal	Susceptible N(%)	Resistant N(%)
Fluconazole	9 (90.00)	1 (10.00)
Itraconazole	9 (90.00)	1 (10.00)
Ketoconazole	9 (90.00)	1 (10.00)
Clotrimazole	8 (80.00)	2 (20.00)
Nystatin	7(70.00)	3(30.00)
Amphotericin-B	10 (100.00)	0(0.00)

Discussion

Identification of causative organisms is important for prescribing appropriate treatment as a wide range of Organisms are isolated in CSOM. The organisms may spread to adjacent structures near the ear or cause local damage to the middle ear itself. Untreated cases of CSOM may result in a broad range of intracranial and extra-cranial complications. Early bacteriological diagnoses of all cases will ensure accurate and appropriate therapy.

In our study out of total 157(100.00%) cases, males 92(58.60%) were more affected than females 65 (41.40%). This finding is in consonance with the study conducted by Ahmed *et* al[(11)] who showed (57.29%) male and (42.70%) female cases out of 192(100.00%) cases in their study. The male predominance may be because of males are more actively involved in outdoor activities, hence more likely to be exposed to contaminated environment.[(12)]

It was observed in present study, In this study the maximum number of cases were found in the age group of 11-20 years 37 (23.57%) followed by 0-10 years 36 (22.93%). These findings were in consistency with a study conducted by O V Akinpelu and Y B Amusa in their study of 160 (100.00%) patients the maximum number of patients were in the age group of 0-10 years 69(43.00%).[(13)]

In the present study 151 (96.18%) specimens were positive and 6 (3.82%) were negative for the culture. The culture results are variable with the other workers also. In present study culture negative could be because of the fact that our hospital is a tertiary care centre. Patient come to us after having sought medical advice from local doctors and having taken multiple or incomplete course of antibiotics. A Srivastava et al studied 112(100.00%) patients with CSOM in 2005. In their study they reported negative culture in 22(19.64%) cases.[(14)]

The most common organism isolated in the present study was Pseudomonas aeruginosa 62(42.17%) followed by Staphylococcus aureus 44(29.93%). These finding is correlated with the other studies conducted by workers like Hiremath S.L. et al (2001)[(15)] and Loy A.C (2002).[(16)]

The most common fungi isolated in present study was Aspergillus niger 8(44.4%). Shrestha SL et al found 6.9% Aspergillus species in their study.[(17)]

Candida albicans 7(38.9%) was the second most common fungi isolated in my study. Kumar H, Seth S found candida albicans in 60% cases in their study.[(2)]

Antibiotic sensitivity was carried out for 147 isolates by Kirby-Bauer disc diffusion method by using antibiotic discs. In the present study 127 (86.4%) of organisms were sensitive to Cefotaxime, followed by Amoxyclav 125 (85%), Amikacin 110 (74.8%), Ciprofloxacin 70 (47.6%), Gentamicin 64 (43.5%), Cotrimaxazole 59 (40.1%) The most effective drugs in the present study are Cefotaxime, Amoxyclav and Amikacin. Similar sensitivity pattern was reported by Shyamala R & Reddy PS.[(18)] However Singh AH et al have found Ciprofloxacin as the most effective drug.[(19)]

Since the resistance to the first commercial antimicrobial agent (penicillin) was identified in 1948,[(20)] almost every known bacterial pathogen has developed resistance to one or more antibiotics in clinical use.[(21)] As antibiotic-resistant pathogens are observed almost concurrently with the use of new antibiotics in hospitals,[(22)] one can easily suppose that wherever antibiotics are used, antibiotic resistance will inevitably follow. Unfortunately, although antibiotic resistance has increased, the development of novel antimicrobial agents has dramatically declined over the past 30 years.[(23)] Therefore, to prevent the return of the pre-antibiotic era, one must use existing antibiotics more judiciously. In present study multidrug resistance isolates were not isolated.

Conclusion

CSOM is major health problem and one of the significant cause of mortality and morbidity around the world, especially in developing countries. This study was conducted with the aim of identification of causative agents associated with ear infections especially in an environment where antibiotics are commonly abused.

Based on the findings from this study, it is therefore recommended that treatment of ear infection is better done when the causative agents as well as the drug sensitivity patterns are known and properly administered Also the role of fungi & anaerobes should be studied. This will enhance better treatment and reduce the burden of the infection on the patients and in the long term, it may reduce the cost of treatment.

It can be concluded that a variety of bacteria are responsible for CSOM with predominance of Pseudomonas aeruginosa followed by Staphylococcus aureus, Klebsiella species, Escherichia coli and Streptococcus pyogenes.

The antibiotic susceptibility testing showed Cefotaxime as the most effective drug followed by Amoxyclav, Amikacin, Ciprofloxacin, Gentamicin and cotrimoxazole.

References

1. Wiwanitkit S, Wiwanitkit V. Pyogenic brain abscess in Thailand. N Am J Med Sci 2012;4:245–8.
2. Kumar H, Seth S. Bacterial and fungal study of 100 cases of chronic suppurative otitis media. J Clin Diagn Res 2011;5:1224–7.
3. World health organization. Chronic Suppurative Otitis Media: burden of illness and management option. WHO Library cataloguing-in-publication data, Geneva, Switzerland 2004.
4. Anwar-us-salam, Abid SH, Abdulla EM. Suppurative Otitis in Karachi: An audit of 510 cases. Pak J Otolaryn 1997;13:66-9.
5. A Srivastava et al. Microbiological Evaluation of an active tubotympanic type of chronic suppurative otitis media. Nepalese journal of ENT Head and Neck Surgery2010;1(2):14-16.
6. Yeo SG, Park DC, Hong SM, Cha C-II, Kim MG. Bacteriology of CSOM — A multicenter study. Acta Oto-Laryngologica 2007;127(10):1062–67.
7. Betty A Forbes, Daniel F Sahm, Alice Weissfeld. Bailey and Scott Diagnostic Microbiology. 12th edition: Elsevier 2007; pp.832-42.
8. Duiguid JP, Collee JG, Fraser AG. Laboratory strategy in the diagnosis of infective syndromes. In: Mackie and McCartney Practical Medical Microbiology (14th Edition), Churchill Livingstone, London 2004.
9. Koneman EW, Allen Stephen D. Colour Atlas and Textbook of diagnostic microbiology. 5th edition. Philadelphia: Lippincott; 1997.
10. Clinical and Laboratory Standard Institute, 2012. Performance standards for antimicrobial susceptibility testing. Clinical and Laboratory Standards Institute, Wayne. 24th Informational Supplement.
11. Ahmed A, Usman J, Hashim R. Isolates from chronic suppurative otitis media and their antimicrobical sensitivity. Pak Armed Forces Med J 1999;49:82-5.
12. Agarwal S.R. Jain A.K. Goyal R. B et al, ―Clinicomycological study of otomycosis with special reference to silent Tympanic Membrane perforation. Ind. J. of Otol. June 2001, Vol. 7, No. 2, 49-52
13. OV Akinpelu et al. Challenges in Management of chronic suppurative Otitis Media in a developing country. The Journal of Laryngology and Otology 2008;122:16-20.
14. A Srivastava et al. Microbiological Evaluation of an active tubotympanic type of chronic suppurative otitis media. Nepalese journal of ENT Head and Neck Surgery2010;1(2):14-16.
15. Hiremath SL, Kanta RC, Yeshwanathrao M, Vasantha Kumar CM. Aerobic bacterial isolates of CSOM and their antibiotic sensitivity pattern. The Indian Practitioner 2001;54(7):486-489.
16. Loy AHC, Tan AL, Lu PKS. Microbiology of chronic suppurative otitis media in Singapore. Singapore Med J 2002;43(6):296-299.
17. Shrestha SL, Amatya RCM, Shrestha I, Ghosh I. Micrbiological Profile of Chronic Suppurative Otits Media. Nepalese journal of ENT Head & Neck Surgery2011;2:6-7.
18. Shyamala R, Reddy PS. The study of bacteriological agents of Chronic Suppurative Otitis Media- Aerobic culture and evaluation. J. Microbiol Biotech Res 2012;2:152-162.
19. Singh AH, Basu R, Venkatesh A. Aerobic bacteriology of Chronic Suppurative Otitis Media in Rajahmundry, Andhra Pradesh, India. Biology & Medicine Journal 2012;4:73-79.
20. Barber M., Rozwadowska-Dowzenko M. Infection by penicillin resistant *staphylococci*. Lancet.1948;2:641–644.
21. Cantas L., Shah S.Q., Cavaco L.M., Manaia C.M., Walsh F., Popowska M., Garelick H., Burgmann H., Sorum H. A brief multi-disciplinary review on antimicrobial resistance in medicine and its linkage to the global environmental microbiota. Front. Microbiol. 2013;4 doi: 10.3389/fmicb.2013.00096.
22. Levy S.B. The challenge of antibiotic resistance. Sci. Am. 1998;278:46–53. doi:10.1038/scientificamerican0398-46.
23. Spellberg B., Powers J.H., Brass E.P., Miller L.G., Edwards J.E., Jr. Trends in antimicrobial drug development: Implications for the future. Clin. Infect. Dis. 2004;38:1279–1286. doi: 10.1086/420937.

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