

Bacteriological Profile of CSOM and Antibiotic Susceptibility Pattern at a Tertiary Care Teaching Hospital, Udaipur, Rajasthan, IndiaSudeepti Saran¹, Viral Chayya², Vikram Singh³, Mohammad Nabil Sindhi⁴, Rambabu Sharma⁵¹PG Resident, Department of ENT, Pacific Institute of Medical Sciences, Udaipur²Professor & HOD, Department of ENT, Pacific Institute of Medical Sciences, Udaipur³Professor, Department of ENT, Pacific Institute of Medical Sciences, Udaipur⁴PG Resident, Department of ENT, Pacific Institute of Medical Sciences, Udaipur⁵Assistant Professor, Department of Microbiology, Pacific Institute of Medical Sciences, Udaipur

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Abstract:**Background:** CSOM (chronic suppurative otitis media) is one of the most important and common global public health problem causing hearing impairment and prevalent particularly in developing countries. Increased irrational and wide use of antibiotics, resistant in the bacterial infection of ear is common.**Objectives:** The main objective of the present study was to appraise the current pattern of bacterial etiology of CSOM in patients and antimicrobial susceptibility of the bacterial isolates prevalent in the hospital.**Materials and Methods:** A prospective observational study was carried out in 125 patients with chronic ear discharge. Sterile swabs were used to collect ear discharge and processed for gram's stain and culture. Antibiotic susceptibility testing was performed by modified Kirby-Bauer disk diffusion method.**Result:** Out of 125 study patients, high prevalence was noted between age group of 21-40 years (41.6%), highest in males (62.4%) as compared to females (37.6%). Among the bacterial isolates, *Pseudomonas aeruginosa* (40.8%), *Staphylococcus aureus* (19.2%), *Proteus* species (6.4%), *Escherichia coli* (10.4%), CoNS (8%), *Klebsiella* species, (4.8%), and *Citrobacter* spp. (3.2%) was the common bacterial isolates. *Staphylococcus aureus* was found to be more sensitive for linezolid and vancomycin. However, the majority of gram-negative isolates showed sensitivity to carbapenems and aminoglycosides.**Conclusion:** *Pseudomonas aeruginosa* was the most common organism isolated on culture followed by *staphylococcus aureus* and *E.coli*. Aminoglycosides, vancomycin and carbapenems had the highest susceptibility rate against the bacterial isolates.**Keywords:** Antibiotic Susceptibility test, Bacterial isolates, CSOM, Ear Discharge, Drug Resistance.This is an Open Access article that uses a funding model which does not charge readers or their institutions for access and distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>) and the Budapest Open Access Initiative (<http://www.budapestopenaccessinitiative.org/read>), which permit unrestricted use, distribution, and reproduction in any medium, provided original work is properly credited.**Introduction**

Chronic suppurative otitis media (CSOM) is an ailment of numerous etiologies and is well known for its persistence and recurrent infections despite the treatment. CSOM is assumed to be a complication of acute otitis media (AOM), but the risk factors are not clear. Chronic suppurative otitis media is a chronic inflammatory process in the spaces of middle ear, results in permanent changes occurs in the tympanic membrane including dimeric membrane formation, atelectasis, perforation, tympanosclerosis, retraction pocket or cholesteatoma [1]. Most commonly buildup of the fluid behind the ear drum, as a consequence of blockage to the Eustachian tube. Other multiple characteristic features seen in CSOM i.e. various degrees of edema, submucosal fibrosis, hypervascularity and infiltration with lymphocyte, plasma and histiocytes [2]. Clinically, CSOM present

in patients with recurrent ear discharge and hearing impairment that may cause serious long-term effects on seeking of language, auditory disturbances, cognitive development and educational process. CSOM is a common disease of all age groups especially of childhood. It is also reported to be one of the major causes of deafness in developing countries as well as in India and Asian subcontinents. Prevalence of CSOM is more common in the lower socioeconomic families because of malnutrition, over-crowding, lack of awareness of personal hygiene, due to inadequate healthcare facilities and frequent infections of the upper respiratory tract [3-4].

Most of the microbiological studies on CSOM have reported that the most common bacterial isolates found in CSOM are *Pseudomonas aeruginosa*,

staphylococcus aureus, proteus mirabilis, E.coli, and Klebsiella species. ³ Pseudomonas aeruginosa, Staphylococcus spp. and some other gram-negative fermenter and non-fermenter organisms such as Proteus species, Escherichia coli, and Moraxella catarrhalis are ubiquitous in nature and present as resident flora on skin and surrounding environment as a normal commensals but in some instances they can easily enter through the perforated ear and cause otitis media [4-5]. Some anaerobes i.e. Bacteroides species, Peptostreptococcus, Propionibacterium species and some opportunistic fungi such as Aspergillus species particularly Aspergillus Niger and Candida species are also responsible for ear infections [6-7]. However, the types of bacterial isolates present in CSOM varies depending on geographical conditions and climate as well.

However, various complications can occur in CSOM if the patients left untreated in the early stage of the disease i.e. persistent otorrhea to mastoiditis, labyrinthitis and facial nerve paralysis, meningitis, thrombosis, and brain abscesses [8]. Thus, management at the early developing stage of CSOM is crucial to avert the threatening complications associated with CSOM. Empirical therapy should be chosen according to the Antibigram pattern of the most common organism i.e. Staph. Aureus, and pseudomonas species associated with CSOM. During recent years, there is decrease complications in CSOM because of early treatment and diagnosis. However, the irrational use of broad spectrum antibiotics has increased the emergence of resistant organisms to the commonly used drugs are very common. This scenario has led to the recurrence and persistence of low-grade infections, which further rise the morbidity rate and financial burden to the patients.

Now days, CSOM has received substantial attention because of its high incidence and chronicity as well as the issues of drug resistance and ototoxicity due to topical and systemic antibiotics. Changes in the microbiological flora and the subsequent advent of sophisticated synthetic antibiotic uses enhance the significance of reappraisal of the contemporary flora in CSOM and in-vitro antibiotic sensitivity pattern has an imperative role for treating doctors to plan a general out-line of treatment for their patients with a chronically discharging ear.

The present deliberation was designed with the aim to find out the local pattern of aerobic microorganisms both (bacterial & fungal) and their antimicrobial sensitivity pattern in patients with CSOM and to provide help to the treating doctors for proper management of the cases for making a protocol for experiential antibiotic therapy.

Therefore, research on bacterial profile and drug susceptibility is essential to aid the treating physician to plan the general management of CSOM and necessary for the ENT surgeon to make the

discharging ear dry through myringoplasty and ossiculoplasty. Nevertheless, the antibiogram of the organisms responsible for CSOM has been found to differ with time and geographical area as well as from one continent to another mostly because of the indiscriminating use of the antibiotics.

Material and methods

Study duration: The present study was carried out for a period of 12 months from January 2022 to December 2022 in the Department of ENT and Microbiology, Pacific Institute of Medical Sciences, Udaipur after obtaining the approval from Institutional Ethical Committee. A total of 125 study participants attending the OPD, ENT department with complaints of ear discharge from >3 months were enrolled in the study. An informed written consent was obtained from all the participants prior to their enrollment in the study.

Objective of the study: The purpose of the present study was to appraise the current pattern of bacteriological profile in CSOM and to determine the antibiotic sensitivity pattern of the aerobic bacterial isolates prevalent in the hospital.

Patient's selection criteria

Inclusion Criteria: Both male and females of any age with discharging ears of unilateral side or bilateral ears and those with ear discharge of >3 months duration were included in the study.

Exclusion criteria: Patients with ear discharge for < 3 months, taking antibiotics currently or preceding 5 days, acute suppurative otitis media, or otitis externa, recent ear surgery, traumatic perforation, any systemic disease, and uncooperative patients were excluded from the study.

Data collection and identification of organisms: A comprehensive history was taken including general examination of ear, nose, throat and neck were conducted thoroughly. Two swabs were taken in each patients of frankly purulent, muco-purulent in nature. Both the swabs were processed immediately in the department of Microbiology. One swab was used for prepared a smear on glass slide for gram's staining. The second swab was used for culture for the isolation of bacteria on culture media such as Blood agar, MacConkey agar for bacterial isolation and Sabouraud dextrose agar were used for isolation of fungal isolates. All the culture plates were incubated at 37° C for 24-48 hours. The plates showing bacterial growth were further examined by based on colony morphology on culture media, gram's stain by colonies on culture media, motility, and biochemical characteristics according to following standard techniques [9]. The isolated bacteria were tested for susceptibility to Penicillin, ampicillin, amikacin, tobramycin, ceftazidime, ceftriaxone, cefotaxime, Amoxclav, cefepime, cefixime, ciprofloxacin, levofloxacin, carbapenems,

doxycycline, gentamicin, vancomycin, ceftioxin, linezolid, cotrimoxazole. ATCC strains (ATCC 25923) used for staphylococcus aureus, (ATCC

25922) for E.coli, and for pseudomonas aeruginosa (ATCC 27853) was used to maintain the quality control in the laboratory. [10-12].



Image 1: MacConkey agar (gram negative bacteria)



Image 2: Blood agar (s. aureus)

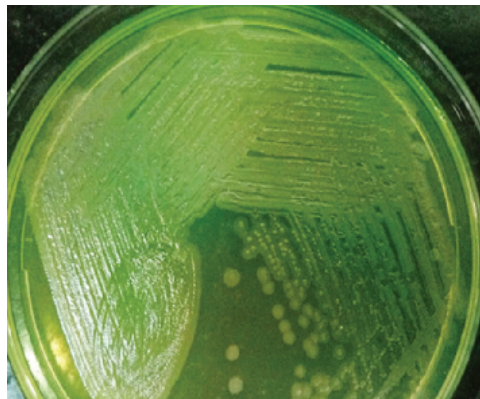


Image 3: Pseudomonas spp.

Results and observations

The present study included 125 patients of CSOM attending in the OPD, Department of ENT at the Pacific Institute of Medical Sciences, Udaipur, Rajasthan. All the subjects were in the age groups ranging from 10 years to 80 years, the highest incidence was found in age group of 21-40 years

(41.6%) followed by 41-60 years (31.2%), >60 years (14.4%) and <20 years (12.8%) as shown in **table 1**. Further, the occurrence of CSOM was found to be higher in males (62.4%) as compared to females (37.6%). Prevalence of unilateral infection was 28.8% in the right ear 48% in the left ear which in turn was more common than bilateral (23.2%) as shown in **table 2**.

Table 1: Distribution of CSOM patients according to age (n=125)

Age groups	No of patients	Percentage (%)
<20 years	16	12.8%
21 – 40 years	52	41.6%
41 – 60 years	39	31.2%
>60 years	18	14.4%

Table 2: Sites of ear infection (n=125)

Site of ear	No of patients	Percentage (%)
Right ear	36	28.8%
Left ear	60	48%
Bilateral	29	23.2%

Among the bacterial growth on culture media, Pseudomonas aeruginosa (non-fermenter) was predominant organism (40.8%), followed by Staphylococcus aureus (19.2%), Escherichia coli

(10.4%), Coagulase negative staphylococcus (8%), Proteus mirabilis (6.4%), Klebsiella species (4.8%), Citrobacter species (3.2%) and Acinetobacter species (2.4%) as shown in **table 3**. Furthermore, among the

fungal isolates, *Candida albicans* (3.2%) was predominant followed by *Aspergillus Niger* (1.6%)

as shown in table 4.

Table 3: Distribution of bacteria causing CSOM (n=125)

Bacterial isolates	Total No.	Percentage (%)
<i>Pseudomonas aeruginosa</i>	51	40.8%
<i>Staphylococcus aureus</i>	24	19.8%
<i>Escherichia coli</i>	13	10.4%
Coagulase negative <i>Staphylococcus</i>	10	08%
<i>Proteus spp.</i>	08	6.4%
<i>Klebsiella spp.</i>	06	4.8%
<i>Citrobacter spp.</i>	04	3.2%
<i>Acinetobacter spp.</i>	03	3.2%

Table 4: Distribution of fungal isolates causing CSOM

Fungal isolate	Total no.	Percentage (%)
<i>Candida albicans</i>	04	3.2%
<i>Aspergillus Niger</i>	02	1.6%

Table 5: Antibiotic susceptibility pattern of gram negative organisms causing CSOM

Name of antibiotics	Name of isolates					
	<i>Pseudomonas spp.</i>	<i>E. coli</i>	<i>Acinetobacter spp.</i>	<i>Klebsiella spp.</i>	<i>Citrobacter spp.</i>	<i>Proteus spp.</i>
Amikacin (30mcg)	91%	98%	92%	93%	97%	96%
Amoxclav (30mcg)	0	100%	92%	88%	93%	91%
Ampicillin+sulbactam(10mcg)	0	91%	89%	84%	91%	89%
Ciprofloxacin (5mcg)	68%	84%	67%	72%	88%	66%
Chloramphenicol (30mcg)	0	95%	82%	83%	87%	69%
Co-trimoxazole (25mcg)	0	87%	66%	78%	68%	73%
Cefepime (30mcg)	77%	79%	63%	64%	64%	64%
Ceftazidime (30mcg)	74%	86%	71%	77%	74%	77%
Cefuroxime (30mcg)	0	91%	69%	82%	78%	62%
Cefotaxime (30mcg)	0	90%	74%	83%	68%	58%
Doxycycline hydrochloride	0	94%	87%	87%	83%	88%
Imipenem (10mcg)	94%	100%	100%	100%	100%	100%
Levofloxacin (5mcg)	76%	78%	88%	84%	88%	83%
Meropenem (10mcg)	97%	100%	100%	100%	100%	100%
Piperacillin+Tazobactam	82%	94%	91%	88%	88%	90%
Tobramycin (10mcg)	91%	96%	92%	91%	93%	92%
Aztreonam (30mcg)	89%	92%	91%	91%	82%	78%
Gentamicin (10mcg)	92%	94%	96%	92%	96%	92%

Pseudomonas aeruginosa was highest sensitive to Meropenem and imipenem (97% & 94%) amikacin and gentamicin (91% & 92%), tobramycin (91%), piperacillin/tazobactam 82%, Aztreonam 89%. less sensitive to ciprofloxacin 68%, ceftazidime 74%, and cefepime 77% as shown in table 5.

All isolates of *Klebsiella* species are sensitive to carbapenems (100%), amikacin 93%, and gentamicin 92%, tobramycin 91% followed by Amoxclav and piperacillin/tazobactam each 88%, ampicillin/sulbactam 84%.

All isolates of *Escherichia coli* was highest sensitive to carbapenems and Amoxclav each 100%, followed by amikacin 98%, tobramycin 96%, gentamicin 94%, doxycycline 94%, ampicillin/sulbactam 91%, piperacillin/tazobactam 94%.

All isolates of proteus species was highest sensitive to carbapenems 100%, amikacin 96%, tobramycin and gentamicin each 92%, Amoxclav 91%, piperacillin/tazobactam 90%, less sensitive to cephalosporins.

All isolates of *Staphylococcus spp.* highest sensitive to amikacin, vancomycin and linezolid was 100%. To add on, their sensitivity to minocycline, gentamicin was higher followed by doxycycline, chloramphenicol and cotrimoxazole. On the other side, they showed less sensitivity against commonly used antibiotics such as clindamycin and erythromycin. Moreover, they were resistant to levofloxacin and ciprofloxacin. **Table 6.**

Table 6: Antibiotic susceptibility pattern of Staphylococcus spp.

Name of antibiotic	Name of isolates	
	<i>Staphylococcus aureus</i>	CoNS
Minocycline (30mcg)	96%	92%
Amikacin (30mcg)	100%	100%
Clindamycin (2mcg)	58%	63%
Cefoxitin (30mcg)	64%	60%
Co-trimoxazole (25mcg)	66%	62%
Ciprofloxacin (5mcg)	24%	18%
Chloramphenicol (30mcg)	91%	97%
Doxycycline hydrochloride (30mcg)	92%	94%
Erythromycin (15mcg)	42%	88%
Gentamicin (10mcg)	94%	93%
Levofloxacin (5mcg)	19%	37%
Linezolid (30mcg)	100%	97%
Vancomycin (5mcg)	100%	98%

Discussion

CSOM is one of the most common and important major health burden in communities leading to hearing impairment with approximately a 5 % global incidence and is particularly prevalent in developing countries. Upper respiratory tract and lower respiratory tract infections, poor hygiene, introduction of foreign body in ear, smoking and misuse of antibiotics were found to be the major risk factors for otitis media. CSOM had several complications associated with this ailment such as irreversible local destruction of middle ear structures facial palsy and serious intracranial and extracranial complications are seen by otologists, pediatricians and general practitioners.

The causative bacteria in CSOM might be aerobic such as *Pseudomonas aeruginosa*, (a ubiquitous pathogen present in hospital environment), *Escherichia coli*, *Staphylococcus aureus*, enterococcus species, *Proteus* spp. and *Klebsiella pneumoniae* as well as *Klebsiella oxytoca* or sometimes anaerobic bacteria's i.e. *Bacteroides* spp., *Peptostreptococcus* and *Propionibacterium* spp. These bacteria are sporadically seen in the skin of the external auditory canal and might multiply in the presence of any injury like trauma, inflammation and in case of high humidity. These bacteria's perhaps gain access into host body by the middle ear through a chronic perforation. Among all these bacterial species, *P. aeruginosa* is the main root cause for deep-seated and progressive destruction of the middle ear and mastoid structures due to releases of its pathogenic toxins and enzymes. [13]

In the current study, out of total 125 CSOM patients, 78 were males (62.4%) and 47 were females (37.6%). The prevalence of unilateral ear involvement (more on the left sided ear) in CSOM patients was found to be more than bilateral sites involvement. Besides, left ear discharge was seen in 48% of the cases while 28.8% of the patients were

with discharge from the right ear and 23.2% of patients had discharge from both ears.

Furthermore, out of 125 culture isolates, 85 isolates were both fermenter and non-fermenter Gram-negative bacteria, 34 isolates were Gram-positive cocci and 6 isolates were fungal agents. The isolate of *Pseudomonas aeruginosa* was found to be more predominant constituting 40.8% (51 isolates) of the isolates and the predominance of *Staphylococcus aureus* was 19.2% (24 isolates) of the total isolates.

Microbiological diagnosis at the early stage of the disease is helpful to ensure rapid and effective treatment to avoid complications in CSOM patients and drug resistance. The current research showed a high prevalence of culture-positive in cases of CSOM. Further, it was seen that CSOM was more prevalent during the first and second decades (upto 40 years) of life and accounting 54% of the CSOM cases. The present findings were in accordance with the observations of previous studies conducted by Kumar and Seth et al., 2011; Bansal et al., 2013 and Rejitha et al., 2014 which reported the maximum number of cases during the second and third decades of life [7,14,15].

In the current study, males were found to be more commonly infected (62.4%) than females (37.6%) and the results of the present study were supported by the findings of Ahmad et al., 1999 [16] where men were 57.3% and women were 42.7%. On the contrary, one of the previous studies carried out by Loy et al., 2002 showed that females are more commonly infected [17]. In general, males were found to be more vulnerable to infection on comparison with females; possibly because of release of androgens hormones in males and estrogens hormones in females modulate host immunity and these sex steroids further affect disease-resistance genes and behavior.

Moreover, amongst aerobic bacterial isolates, the predominance organism isolated of *Pseudomonas*

aeruginosa was found to be higher (40.8%) followed by *S. aureus* (19.2%), *E. coli* (10.4%), *Coagulase-negative Staphylococcus* (8%), *Proteus species* (6.4%), *Klebsiella species* (4.8%), *Citrobacter species* (3.2%) and *Acinetobacter species* (2.4%). Besides, *Aspergillus species* (1.6%) was the predominant fungus followed by *Candida species* (3.2%). Together *Staphylococcus aureus* and *Pseudomonas species* account for more than 60% of cases. Similar findings were observed in studies conducted by Shyamala et al. Iseh and Adegbite et al., [18,19,20]. But in contrast of the disease Loy et al. 2002 reported of *Staphylococcus aureus* as the major causative agent of ear infection and present in CSOM patients [17]. Several researches from other different nations including India, Nepal, Nigeria and Singapore have shown that *P. aeruginosa* is the most common pathogen responsible for CSOM, followed by *S. aureus* Yeo et al. [21], Madana et al. reported 32%, [22] Afolabi et al. reported 31.3% [23, 24]. Whereas, other counter studies conducted in other countries such as Saudi Arabia, Pakistan, and Iran have demonstrated that *S. aureus* is the predominant pathogen in CSOM patients, followed by *P. aeruginosa*. Mariam et al. reported as 65.2% prevalence of *pseudomonas aeruginosa* in her study [25]. This difference of bacterial isolation rate in CSOM patients might be possible due to an association with climate effect and geographical distribution.

After bacterial isolation, the antibiotic susceptibility testing and their sensitive pattern was tested for all the organisms and according to present findings, *P. aeruginosa* antibiotics susceptibility pattern showed high sensitivity to Meropenem, imipenem, tobramycin, gentamicin, amikacin, Aztreonam, piperacillin-tazobactam followed by cefepime and levofloxacin as shown in table 5. Though, their sensitivity was less for fluoroquinolones and least for ceftazidime in the current study. The present results support the findings of a previous study carried out by Harshika YK et al. in which *Pseudomonas aeruginosa* were highly susceptible to imipenem and amikacin (93.6%) followed by gentamicin (89.3%) [26]. Any defect in the tympanic membrane from an acute episode of otitis media might lead to the invasion of *Pseudomonas species* to the middle ear.

Moreover, according to present findings, gram-negative bacteria apart from *Pseudomonas* showed 100% sensitivity to carbapenems; followed by highly sensitive to amikacin, gentamicin and tobramycin, moderate sensitivity to amoxiclav, ampicillin-sulbactam ciprofloxacin, levofloxacin, co-trimoxazole and, doxycycline. Though, the sensitivity was less for third-generation cephalosporins as shown in table 5. Similar results were shown by the study of Worku and Bekele [27]. Further, amikacin (90%) and gentamicin (89%) have shown good results for both gram-positive and gram-negative pathogens [28].

Staphylococcus aureus was found to be highly susceptible for the majority of the antibiotics except for Penicillin, the sensitivity of the isolates was found to be 100% for amikacin, vancomycin and linezolid, followed by good sensitivity to gentamicin (94%) and minocycline (96%), doxycycline (92%), chloramphenicol (91%) and least sensitive to levofloxacin (19%) and cephalosporins. All the 24 isolates of *Staphylococcus aureus* were Methicillin Sensitive *Staphylococcus aureus* (MSSA). The present findings were contradictory to the expectation in the current scenario where misuse of antibiotics results in the rapid increase in infections caused by MRSA. Numerous previous studies which supported present findings are Agrawal et al., 2014; Chaudhary, et al., 2014; Sattar Abdul et al., 2012; and Singh et al., 2012 [29-31]. The high isolation of *Staphylococcus aureus* followed by *pseudomonas aeruginosa* in CSOM patients might be the result of ubiquitous nature as well as the higher rate of colonization in the auditory canal and upper respiratory tract. In the current study, the resistant pattern towards most commonly used topical and oral antibiotics in cases of CSOM is perhaps because of an indiscriminating intake of antibiotics.

Otitis media has an imperative role in causing hearing impairment and the situation is worrisome, especially in the pediatric age group. In case of delayed management, it may result in serious complications later in life in language development, communication skill and educational process. In the present scenario, in patients with CSOM, both microbial profile and their antimicrobial susceptibility pattern change from time to time might be because of geographical and environmental variations in the study population. [32] As the strains of bacterial isolates responsible for CSOM are still found to be responsive to first line drugs in our study area. The treatment of chronic suppurative otitis media should be tailored according to the pattern in the microbiological flora of each discharging ear. More such studies are required in this context, as they will help to the clinicians in selecting the optimum preemptive treatment therapy in the CSOM cases.

Conclusion

From current research, it has been speculated that the majority of the organisms showed resistance towards regularly used cell wall inhibitors such as penicillin group of drugs and cephalosporins. Thus, the clinician should have awareness of the fact that the antibiotic susceptibility pattern of the CSOM cause organisms to keep changing and this variation is got worsen with inappropriate use of antibiotics, which is capable of creating multidrug resistance among the organisms; thus making the management of CSOM even more difficult. The findings of the present research will immensely contribute to the effective management of CSOM. Furthermore, a future study

can be planned to enquire if the bacteriological profile of microorganisms and their antibiotic sensitivity pattern are affected by the size and chronicity of perforation as well as the character of drainage. Besides, regular monitoring and follow up of the patients are more effective. Continue monitoring will assist in developing an antibiotic policy in the hospital which in turn will be useful in promoting the rational use of antibiotics and preventing the emergence of resistance.

References

1. Acuin J. Global burden of disease due to chronic suppurative otitis media: Disease, deafness, Deaths and DALYs Chronic Suppurative Otitis media-Burden of illness and Management Options. Geneva: World Health Organization;2012; 9-23.
2. Probst R, Grevers G, Iro H et al, Basic Otorhinolaryngology. 2006;166- 197.
3. K Sanjay, Sharma R, Saxena A, Panndey A, Gautam P, Taneja V. Bacterial Flora of Infected Unsafe CSOM. 2012, Indian Journal of Otolaryngology; 2012;18(4): 288-211.
4. Kelvin Kong, Harvey, L.C. Otitis Media 2009: An Update-Natural History, definitions, risk factors and burden of otitis media. Med J Aust. 2009;191: S39S43.
5. Verhoeff, M., van der Veen, E. L., Rovers, M., Sanders, E.A., Schilder et al, Chronic suppurative otitis media: a review. Int J Pediatr Otorhinolaryngol. 2006;70: 1-12.
6. Poorey, V.K., Arati, Iyer. Study of Bacterial Flora in Chronic Suppurative Otitis Media and its clinical significance. Ind. J. Otolaryngol H & N Surg. 2002;54(2):91- 95.
7. Kumar H, Seth S. Bacterial and Fungal Study of 100 cases of Chronic Suppurative Otitis Media., Journal of Clinical and Diagnostic Research. 2011; 5(6):1224-1227
8. Tomasz, A., Multiple-antibiotic resistant pathogenic bacteria. A Report on the Rockefeller University Workshop. New Engl-J Med. 1994; 330:1247-1251.
9. Collee, J. G., Duguid, J. P., Fraser, A.G., et al, Laboratory strategy in the diagnosis of infective syndromes. Mackie and McCartney practical medical microbiology, 14th edn. Churchill Livingstone, Singapore. 1996; 53 - 94.
10. CLSI, Performance standards for antimicrobial susceptibility testing; twenty-fourth informational supplement. CLSI document M100- S24. Clinical and Laboratory Standards Institute, Wayne, P.A. 2019.
11. CLSI. Performance standard for antimicrobial susceptibility testing; Twenty-fourth informational supplement. Wayne, PA. Clinical and Laboratory standard Institute. 2014;34 (1) CLSI document M100-S24.
12. Basu S, Pal A, Desai PK. Quality control of culture media in a microbiology laboratory. 2005, Indian J Med Microbiol. 23(3):159-63.
13. Acuin J. Chronic Suppurative Otitis Media- Burden of Illness and Management Options. Geneva: World Health Organisation; 2004. Available from: http://www.who.int/pbd/publications/Chronicsuppurativeotitis_medi a.pdf.
14. Bansal Sulabh, Ojha Tarun, Kumar Suresh, Singhal Amit, Vyas Pratibha. Changing Microbiological trends in cases of CSOM. Int J Cur Rev. 2013; 5(15): 76-81
15. Rejitha, I. M., Sucilathangam, G., Kanagapriya, M., Microbiological Profile of CSOM in a Tertiary Care Hospital. International Journal of Scientific Research. 2014; 3 (2): 474 475.
16. Ahmed A, Usman J Hashim R. isolates from chronic suppurative otitis media and their antimicrobial sensitivity. Pak Armed forces Med J. 1999; 49:82-5.
17. Loy AH, Tan AL, Lu PK. Microbiology of chronic suppurative otitis media in Singapore. 2002, Singapore Medical Journal;43(6):296-9.
18. Shyamla R, Reddy SP. The study of bacteriological agents of chronic suppurative otitis media-aerobic culture and evaluation. J Microbiol Biotechnol Res. 2012;2:152-62.
19. Iseh KR, Adegbite T. Pattern and bacteriology of acute suppurative otitis media in Sokoto, Nigeria. Ann Afri Med. 2004; 3: 164-6.
20. Aslam MA, Ahmed Z, Azim R. Microbiology and drug sensitivity patterns of chronic suppurative otitis media. J Coll Physicians Surg Pak. 2004;14:459-61
21. Yeo SG, Park DC, Hong SM, et al. Bacteriology of chronic suppurative otitis media-a multicenter study. Acta Otolaryngol. 2007; 127 (10):1062-7.
22. Madana J, Yolmo D, Kalaiarasi R, et al. Microbiological profile with antibiotic sensitivity pattern of cholesteatomatous chronic suppurative otitis media among children. Int J Pediatr Otorhinolaryngol; 2011;75 (9):1104-8.
23. Afolabi OA, Salaudeen AG, Ologe FE, et al. Pattern of bacterial isolates in the middle ear discharge of patients with chronic suppurative otitis media in a tertiary hospital in North central Nigeria. Afr Health Sci. 2012;12 (3):362-7.
24. Afolabi OA, Aremu SK, Alabi BS, et al. Traumatic tympanic membrane perforation: an aetiological profile. BMC Research Notes. 2009; 2:232.
25. Mariam KA, Ahsanullah M, Mehtab J, et al. Prevalence of bacteria in chronic suppurative otitis media patients and their sensitivity patterns against various antibiotics in human population of Gilgit. Pak J Zool. 2013;45(6):1647-53
26. Harshika YK, Sangeetha S, Prakash R. Microbiological profile of CSOM and their antibiotic sensitivity pattern in a tertiary care hospital., Int J Curr Microbiol App Sci. 2015; 4(12):735-43.

27. Worku M, Bekele M. Bacterial isolate and antibacterial resistance pattern of ear infection among patients attending at Hawassa university referral Hospital, Hawassa, Ethiopia. *Indian J Otol.* 2014;20(4):155
28. Archana Garg, Lakshmi Agarwal, Mamta Gupta, Rishabh Mathur et al, A Study on Bacteriological Profile and the Antibiotic Susceptibility Pattern in Cases of Chronic Suppurative Otitis Media in Haroti Region, *Medical Journal of Dr. D.Y. Patil Vidyapeeth.* Published by Wolters Kluwer – Medknow. 2021.
29. Agrawal A, Kumar D, Goyal A, Goyal S, Singh N, Khandelwal G, et al, Microbiological Profile and their Antimicrobial Sensitivity Pattern in patients of otitis media with ear discharge. *Indian J Otol.* 2013; 19(1):5-
30. Chaudhary, B. L., Snehanshu Shukla. Bacteriological Profile and their Antibiotic Susceptibility pattern in cases of otitis media. *Bulletin of Pharmaceutical & Medical Sciences.* 2014; 2 (2):2209-2212.
31. Abdul Sattar, Alamgir, A., Hussain, Z., Sarfraz, S., Nasir, J., Badar-e-Alam et al, Bacterial Spectrum and their Sensitivity Pattern in Patients of Chronic suppurative otitis media. *Journal of the College of Physicians and Surgeons Pakistan.* 2012;22 (2):128 129
32. Prakash R, Juyal D, Negi V, Pal S, Adekhandi S, Sharma M, et al. Microbiology of chronic suppurative otitis media in a tertiary care setup of Uttarakhand state, India. *N Am J Med Sci.* 2013; 5(4):282-87.