

Antibiogram of chronic suppurative otitis media in relation to the current bacteriological profile in a tertiary care hospital

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ABSTRACT

Background: Every year, about 31 million cases of chronic suppurative otitis media are reported. Due to a lack of access to healthcare, inadequate sanitation and hygiene standards, a lack of knowledge about ear care, underlying socioeconomic issues, and a lack of infrastructure for diagnosis and treatment, it is a common illness in low-income countries. The objective of this study is to determine the bacterial profile and susceptibility pattern of microorganisms isolated from chronic suppurative otitis media patients.

Material and Methods: This Descriptive cross-sectional study was conducted at the Department of Microbiology, Izzat Ali Shah Hospital and POF Hospital Wah Cantt from December 2022 to January 2024. A total of 105 ear swabs were received for culture and sensitivity. Growth was observed in 70 ear swab cultures after 48 hours of incubation at 35±2°C at ambient air. Isolates were identified as *Staphylococcus aureus*, *Enterococcus spp*, *Enterobacterales* including *E. coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, and *Pseudomonas aeruginosa*. Kirby Bauer disk diffusion method was used to assess susceptibility profiles of positive cultures using Muller Hinton agar incubated at a temperature of 35±2°C for 24 hours in ambient air.

Results: Out of all 70 positive cultures, 40 isolates were gram-positive cocci while 30 isolates were gram-negative rods. In gram-positive isolates, *Staphylococcus aureus* was 90% susceptible to ampicillin while *Enterococcus spp*. was found to be 99% susceptible. Ampicillin was not tested for gram-negative organisms because of its proven inactivity against gram-negative bacteria. Susceptibility of *Staphylococcus aureus* was also tested against Penicillin and cefoxitin, where it was found to be 95% and 90% sensitive, respectively. Vancomycin was tested by minimum inhibitory concentrations using E-strip method. No resistance was observed in *Staphylococcus aureus* against vancomycin however, 0.5% vancomycin-resistant enterococcus spp were observed. *Pseudomonas aeruginosa* and *Enterobacterales* were found to have variable susceptibility to antimicrobials tested including Ceftazidime, Gentamicin, Imipenem, Ceftriaxone, cefepime, and Ciprofloxacin.

Conclusion: Bacterial profile and susceptibility pattern of microorganisms isolated from chronic suppurative otitis media should be carried out routinely which can help establish an antibiogram as well as guide empirical therapies.

Keywords: Antibiogram, Antibiotic stewardship, CSOM, Ear swab culture

BACKGROUND

Chronic suppurative otitis media (CSOM) manifests as persistent irritation of the middle ear, accompanied by tympanic membrane perforation and continuous ear

discharge. These symptoms can persist for up to six weeks, highlighting the chronic nature of the condition.¹ Chronic otitis media with effusion (OME) is characterized by the presence of middle ear effusion without acute signs of infection. While OME may originate from acute otitis media, it differs in its chronicity and lacks the acute inflammatory manifestations. This condition is often associated with conductive hearing loss due to the accumulation of fluid in the middle ear space.²

Approximately, 31 million CSOM cases are encountered per year. It's a prevalent illness in nations with little resources due to limited access to healthcare, poor hygiene and sanitation practices, lack of education on ear care, underlying socioeconomic factors, and inadequate infrastructure for diagnosis and treatment.^{3,4}

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Children under the age of two are most susceptible to CSOM due to a short ear canal, however, older children and adults have been found to have higher rates of the condition.^{5,6}

There are two variants of CSOM. Attico-antral type and Tubo-tympanic type.⁷ Microorganisms such as *Staphylococcus aureus*, CONS, *Pseudomonas aeruginosa*, *Proteus mirabilis*, *Enterococcus spp.*, and others are frequently isolated.⁸ The symptoms of CSOM include middle ear discharge that usually lasts longer than six weeks and hearing loss brought on by tympanic membrane perforation.⁹ CSOM is considered the most common cause of preventable hearing loss. Extracranial and intracranial issues might result in severe, sometimes fatal side effects from insufficiently treated CSOM.

Analyzing the bacteriological profile of microbes causing CSOM and their susceptibility to existing antibiotics is crucial for guiding effective treatment strategies. By identifying the specific pathogens responsible for CSOM and understanding their antibiotic susceptibility patterns, clinicians can tailor treatment plans to target the causative organisms more effectively. This approach not only improves treatment outcomes but also helps mitigate the risk of antibiotic resistance and the development of complications associated with inadequately treated CSOM. Through our analysis, we aim to provide valuable insights into the microbial landscape of CSOM and inform evidence-based antibiotic prescribing practices for better patient care.

MATERIAL AND METHODS

This descriptive cross-sectional study was carried out at the Department of Microbiology, Izzat Ali Shah Hospital Wah Cantt, from December 2022 to January 2024. We determined a sample size of 105 by keeping the prevalence of CSOM is 275.⁴ After approval of the IRB, informed consent was obtained from all the patients included in the study.

Patients who had not received any systemic or local antibiotics for seven days before sample collection and who experienced unilateral, bilateral, chronic, or intermittent ear discharge through a perforated tympanic membrane for more than 12 weeks duration were included in the study. Duplicate samples, patients taking antibiotics before sample collection and patients with acute suppurative otitis media, chronic otitis media with effusion, and otitis externa were excluded from the

study. Contaminated samples yielding mixed growths were also excluded from the study. With a sterile cotton-tipped swab stick, two aseptic aural swabs were taken from each patient's discharging ear, one for microscopy and the second for bacterial culture. After the auditory canal was cleared with 70% ethanol, a sterile aural speculum was used to direct insertion of a swab stick into the middle ear to collect pus without coming into contact with the surrounding skin. The sterile tubes containing swabs were transported to microbiology lab as soon as possible and processed according to lab protocols.

Microscopy: A uniformly thin smear was prepared on a sterile glass slide from each swab. It was lightly flame-fixed and then air-dried. Gram staining was performed and the smear was then examined under oil immersion (x100) lens for presence of any pus cells and identifiable microorganisms.

Culture and Identification of Bacteria: The swab was inoculated onto Blood and MacConkey agar plates (Oxoid, UK) and incubated in ambient air at 37°C for 24-48 hours. Any bacterial growth was identified using morphological characteristics of the growth and microscopy (gram reaction, shape, organization). Gram-positive isolates were subjected to catalase and coagulase tests to differentiate between different *Staphylococcus* and *Enterococcus spp.* Gram-negative isolates were further identified by lactose fermentation properties, oxidase, citrate utilization, motility, indole, urease, and triple sugar iron tests. Antibiotic susceptibility of isolated bacterial pathogens was evaluated using the modified Kirby Bauer disc diffusion method, in compliance with the guidelines provided by the Clinical and Laboratory Standard Institute (CLSI) 2024.¹⁰ For each isolate, a 0.5 McFarland standard solution was prepared in sterile normal saline by mixing a loopful of well-isolated colonies, taken from blood agar plate after a growth of at least 18-24 hours. Mueller-Hinton agar (Oxoid, UK) was lawned by dipping a sterile swab in a prepared solution. A variety of commercially available antibiotic discs (Oxoid, UK) were placed on the media and incubated at 37°C in ambient air for 24 hours. According to CLSI criteria, the results were recorded as resistant and sensitive after observing zone diameter formed around all the tested antibiotic discs. Gram-positive organisms were tested for susceptibility against the following antibiotics: amoxicillin/clavulanate (20/10 µg), cefoxitin (30 µg),

ampicillin (30 µg), ceftriaxone (30 µg), clindamycin (02 µg), ciprofloxacin (05 µg), gentamicin (10 µg), imipenem (10 µg), linezolid (30 µg), and vancomycin (30 µg). Gram-negative isolates were tested for susceptibility against amoxicillin/clavulanate (20/10 µg), Ceftazidime (10 µg), Gentamicin (10 µg), Imipenem (10 µg), Ceftriaxone (30 µg), cefepime (30 µg), and Ciprofloxacin (5 µg). *Escherichia coli* ATCC 25922 and *Staphylococcus aureus* ATCC 25923 were used as QC strains for agar medias as well as for antimicrobials.

Data was analyzed using statistical package for social sciences, SPSS version 23. Frequencies and percentages were calculated for categorical variables. Chi-square test used as the test of significance keeping a significant p-value of <0.05.

RESULTS

A total of 105 ear swab samples were included in the study. Gender distribution of all included patients is shown in Table-I.

Out of 105 cultured samples, 70 yielded pure growth of various microorganisms. 25 samples did not yield any

growth after 48 hours of incubation while 10 samples yielded contaminated growth. *Pseudomonas aeruginosa* and *S. aureus* were the predominant isolates (30% and 20% respectively). Other isolated organisms included Enterococcus spp, Coagulase-negative staphylococcus spp. (CONS) and Enterobacterales including Klebsiella spp., Proteus spp., and *E. coli*. Frequency and percentages of all organisms isolated is shown in Table-II. The Antibiotic sensitivity profile of microorganisms is shown in Table-III. *P. aeruginosa* was found to be most sensitive to Tazobactam-piperacillin (TZP) i.e. > 90%. Amikacin was the most effective antibiotic observed after carbapenems. However, Gentamicin was not found to be as susceptible as Amikacin. Sensitivity to Ciprofloxacin and Ceftazidime was also observed to be low. *S. aureus* demonstrated ≥90% sensitivity to both Vancomycin and linezolid where Vancomycin was tested by E-strip while Linezolid was tested by disc diffusion method. Cefoxitin disc diffusion method was used to differentiate between Methicillin-sensitive (MSSA) and Methicillin-resistant *Staphylococcus aureus* (MRSA). About 95% of isolates were MSSA.

Table-I: Gender distribution of patients (n=105).

Gender	n (%)
Female	34 (32.39%)
Male	71 (67.61%)
Total	105

Table-II: Percentages and frequencies of isolated organisms (n=70).

CSOM EAR SWABS	Microbiota n (%)
<i>Staphylococcus aureus</i>	30(42.85%)
<i>Enterococcus</i> spp.	5(7.1%)
Enterobacterales (<i>E. coli</i> , <i>K. pneumoniae</i> , <i>Enterobacter cloacae</i> , <i>Proteus mirabilis</i>)	10(14.28%)
<i>Pseudomonas aeruginosa</i>	20(28.57%)
CONS	5(7.1%)
Total	70(100.0%)

Table-III: Antibigram of CSOM (% susceptible).

Antimicrobials	<i>Staphylococcus aureus</i> & CONS	<i>Enterococcus</i> spp	Enterobacterales	<i>Pseudomonas aeruginosa</i>
Ampicillin	40%	98%	Not tested	Not tested
Cefoxitin	95%	-	Not tested	Not tested
vancomycin	100%	99%	Not tested	Not tested
linezolid	100%	100%	Not tested	Not tested
Erythromycin	90%	-	Not tested	Not tested
Gentamycin	90%	Not tested	80%	87%
ciprofloxacin	100%	100%	80%	60%
amikacin	Not tested	100%	100%	100%
Piperacillin-tazobactam	-	-	-	100%
Meropenem	-	-	100%	100%
ceftriaxone	-	-	100%	Not tested
Trimethoprin-sufamethoxazole	100%	-	100%	Not tested

DISCUSSION

The majority of studies on similar topics regard *S. aureus* to be the most prevalent bacterium. According to our research, *S. aureus* is also the most prevalent isolate. The second-most prevalent isolate in this study was *P. aeruginosa*. *P. aeruginosa* colonizes the auditory canal more frequently as compared to other bacteria due to several advantageous features, including a low nutritional need for life and relative resistance to antibiotics and their byproducts, and the ability to form biofilm. The organism is shielded from the typical host defense systems and antibiotic drugs by the devitalized tissue, disrupted circulation, and damaged epithelium that make up the niche. The organism also thrives in the external auditory canal, functions as an opportunistic pathogen, and may result in suppurative illness in adjacent areas¹¹ Enterobacterales including *Citrobacter spp.*, *Enterobacter spp.*, *Proteus spp.*, *E. coli*, and *Klebsiella spp.* were among the other bacterial isolates. Gram-positive isolates other than *Staphylococcus aureus* included CONS and Enterococcus spp. Similar growth patterns were observed in several other studies conducted in Bangladesh and China.^{14,15} Growth of polymicrobial organisms was also frequent.^{12, 13}

Because of bacterial, environmental, and host variables, CSOM is most likely a complex illness. Bacteria from the external auditory canal that enter through a ruptured tympanic membrane can induce CSOM.¹⁶ Use of contaminated water for bathing/showering may also play a role in pathogenesis of CSOM.¹⁶⁻¹⁸ Pathogenic factors leading to CSOM may include bacterial biofilm formation.¹² Additional variables include increased proinflammatory cytokines, reduced middle ear ciliary activity, and overproduction of mucin.^{18,19}

The antibiogram analysis of chronic suppurative otitis media (CSOM) in our research study revealed *Staphylococcus aureus* susceptibility to Ampicillin and Penicillin at 90%, and to cefoxitin at 95%, with 100% sensitivity to all other antimicrobials tested. For Enterococcus, susceptibility to ampicillin and penicillin was 99%, while vancomycin exhibited 99.5% sensitivity, with all other antibiotics showing complete sensitivity. Enterobacteriaceae and Pseudomonas demonstrated susceptibility to all antimicrobials in the antibiogram. This study's findings were compared with a similar investigation conducted in Pakistan and another in Malaysia.^{20,21}

Timely and appropriate treatment of upper respiratory tract infections (URTI) is paramount to prevent the progression to community-acquired severe otitis media (OSOM). Incomplete courses of therapy contribute to the emergence of resistant organisms, underscoring the importance of completing prescribed treatments. Physicians should prescribe antibiotics with broad-spectrum coverage, targeting both Gram-positive and Gram-negative bacteria, along with anaerobic organisms. Accessible culture results are essential for accurate antibiotic de-escalation. In regions where first-line antibiotic resistance is prevalent due to irrational and excessive antibiotic use, last-resort medications may be the only viable treatment options. Therefore, promoting awareness and adherence to appropriate antibiotic stewardship practices is crucial in combating antimicrobial resistance and improving patient outcomes in URTI and associated conditions like OSOM.

CONCLUSION

The significant predominance of microbiota in the bacteriological profile of chronic suppurative otitis media (CSOM) underscores the challenge of antibiotic susceptibility. To address this, it's crucial to stay abreast of recent shifts in bacteriological profiles and tailor antibiotic prescriptions based on sensitivity patterns. Utilizing local antibiograms for antibiotic selection is paramount, with a focus on judicious antibiotic use. Regular assessments of microbiological profiles and antibiotic sensitivities are necessary to develop and update local antibiograms, providing valuable empirical guidance in clinical practice.

CONFLICT OF INTEREST

None

GRANT SUPPORT & FINANCIAL DISCLOSURE

Declared none

AUTHOR CONTRIBUTION

Naila Iqbal: Main idea

Saba Anwar: Data collection

Saira Salim: Data analysis

Tahira Tehseen: Result writing

Saleha Ali: Discussion writing

Asif Raza: Proof reading

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