Antimicrobial Susceptibility Pattern of Gram-Negative Bacterial Isolates in Chronic Mucosal Otitis Media

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ARTICLE INFO

Received: 5 August 2026
Accepted: 12 August 2026
Published Online: 19 August 2026

DOI: 10.5281/zenodo.22012088

Volume: 10, Number: 1, Page: 185-188

e-ISSN: 2789-5912
ISSN: 2617-0817

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ABSTRACT

Background: Chronic mucosal otitis media is a long-lasting middle ear infection in children from developing nations, primarily due to Gram-negative bacteria, particularly Pseudomonas aeruginosa, which shows rising resistance to ampicillin and cephalosporins, whereas ciprofloxacin, aminoglycosides, and carbapenems continue to be more effective. Insufficient information in Bangladesh indicates a necessity for improved surveillance. This study assesses the antimicrobial resistance of Gram-negative isolates in a tertiary care facility. **Methods & Materials:** This cross-sectional research conducted at Dhaka Medical College Hospital, Bangladesh (2019 and 2022–2023) involved patients between the ages of 6 and 55 with chronic mucosal otitis media. Aural swabs were cultured on blood agar and MacConkey agar, and isolates were identified using standard techniques. Antimicrobial susceptibility was assessed through the Kirby–Bauer disk diffusion technique, followed by descriptive statistical analysis conducted after obtaining ethical approval and informed consent. **Results:** Out of 83 isolates, 58 (69.9%) were Gram-negative, predominantly Pseudomonas aeruginosa (62.1%), and including Klebsiella, E. coli, and Proteus. P. aeruginosa exhibited the greatest sensitivity to gentamicin and piperacillin-tazobactam but demonstrated significant resistance to ciprofloxacin and cephalosporins. Other isolates demonstrated higher susceptibility to meropenem, amikacin, and piperacillin–tazobactam while showing significant resistance to ampicillin. **Conclusion:** Chronic mucosal otitis media was primarily dominated by gram-negative bacteria, especially Pseudomonas aeruginosa, which showed significant resistance to standard antibiotics; however, carbapenems, aminoglycosides, and piperacillin–tazobactam proved to be the most effective treatments. Therapy and monitoring guided by culture are essential.

Keywords: *Antimicrobial Susceptibility Pattern, Gram-Negative Bacterial, Chronic Mucosal Otitis Media*

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INTRODUCTION

Chronic Mucosal Otitis Media (CMOM) is a lasting ear infection characterized by ear discharge, primarily impacting children and prevalent in developing nations, linked to inadequate socioeconomic conditions [1]. Approximately 31 million individuals experience CMOM each year globally, with significantly higher rates in developing nations—up to three times compared to developed areas [2,3].

CMOM is primarily induced by aerobic Gram-negative bacteria, with Pseudomonas aeruginosa being the predominant pathogen. Other commonly isolated organisms consist of Enterobacteriaceae like Klebsiella, Proteus, Escherichia coli, and Citrobacter spp., whereas Staphylococcus aureus is the prevalent Gram-positive isolate [4,5]. Pseudomonas aeruginosa is the predominant pathogen in CSOM, continuing to thrive because of its toxin generation, ability to endure wet environments, and biofilm development that shields it from immune responses and antibiotics, leading to persistent infection and unsuccessful treatment [6]. Antimicrobial resistance in CMOM is rising, leading to diminished effectiveness of frequently used antibiotics such as ampicillin, amoxicillin, and cephalosporins. Fluoroquinolones and aminoglycosides continue to show greater effectiveness against Gram-negative isolates, although resistance is developing [7,8].

Research indicates that Pseudomonas aeruginosa and other Gram-negative bacteria exhibit differing antibiotic susceptibility by location, showing significant sensitivity to ciprofloxacin and aminoglycosides, whereas resistance to β-lactams is rising [9]. The production of extended-spectrum β-lactamases (ESBL) in Gram-negative bacteria has complicated the treatment of CSOM, since strains that produce ESBL tend to be multidrug-resistant, which reduces available therapeutic options and heightens the risk of treatment failure [3].

Studies from various regions indicate differing antimicrobial susceptibility in CSOM isolates, demonstrating significant resistance to medications such as amoxicillin-clavulanate and third-generation cephalosporins, while ciprofloxacin and amikacin typically continue to be effective [10,11].

Studies conducted in Bangladeshi tertiary hospitals on CSOM revealed a predominance of Gram-negative bacteria, primarily Pseudomonas aeruginosa, along with Klebsiella, Proteus, and E. coli. Increased resistance was observed to ampicillin, amoxicillin, and cephalosporins, while enhanced sensitivity was noted for ciprofloxacin, gentamicin, and amikacin. Carbapenems continued to be the most effective against resistant isolates, highlighting the significance of antibiotic selection based on culture [12,13].

In Bangladesh, the information regarding Gram-negative bacteria and antimicrobial resistance in mucosal CSOM is scarce and inconsistent, obstructing effective empirical therapy and leading to increased resistance. The research seeks to identify the pattern of antimicrobial susceptibility in Gram-negative isolates from chronic mucosal otitis media at a tertiary care facility in Bangladesh.

METHODS & MATERIALS

Study Design and Setting

This cross-sectional observational study was conducted in the Department of Otolaryngology and Head–Neck Surgery, Dhaka Medical College Hospital, Dhaka, Bangladesh. The study was carried out during two data collection periods: from March 1, 2019 to September 30, 2019, and from March 1, 2022 to February 28, 2023.

Study Population and Eligibility Criteria

Patients attending the outpatient and inpatient services of the Department of Otolaryngology and Head–Neck Surgery with a clinical diagnosis of chronic mucosal otitis media (CMOM) were considered for enrollment. Patients aged 6–55 years of either sex who provided informed consent were included in the study.

Patients with otomycosis, otitis externa, or other forms of otitis media were excluded. In addition, patients who had received systemic or topical antibiotics within the preceding seven days were not included to avoid alteration of bacterial growth and antimicrobial susceptibility patterns.

Clinical Evaluation and Specimen Collection

A detailed history was obtained from each participant, followed by thorough otorhinolaryngological and relevant systemic examinations. After clinical confirmation of CMOM, aural discharge specimens were collected aseptically from the affected ear using sterile cotton swabs under direct visualization with a sterile aural speculum. Care was taken to avoid contamination from the external auditory canal and surrounding structures.

The collected specimens were immediately transported to the microbiology laboratory for culture and sensitivity testing.

Isolation and Identification of Bacterial Isolates

The specimens were inoculated onto blood agar and MacConkey agar media and incubated aerobically at 37°C for 24–48 hours. Following incubation, culture plates were examined for bacterial growth. Colony characteristics were assessed, and isolates were identified using Gram staining, colony morphology, hemolytic characteristics on blood agar, motility testing, and standard biochemical tests according to established microbiological procedures.

Pure bacterial isolates were selected for further analysis. Organisms were categorized as Gram-positive or Gram-negative based on Gram-staining characteristics.

Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing was performed using the Kirby–Bauer disk diffusion method on Mueller–Hinton agar following standard laboratory guidelines. A panel of commonly prescribed antibiotics used in the treatment of otologic infections was tested. Following incubation, zones of inhibition were measured and interpreted as susceptible or resistant according to standard interpretative criteria.

The antimicrobial susceptibility profiles of Gram-negative bacterial isolates, including *Pseudomonas aeruginosa, Escherichia coli, Klebsiella pneumoniae*, and *Proteus* species, were analyzed and recorded. Antibiotics not routinely tested against specific organisms were reported as not tested (NT).

Data Collection and Quality Control

Clinical, microbiological, and antimicrobial susceptibility data were recorded using a structured data collection form. Standard microbiological procedures were followed throughout specimen collection, culture, organism identification, and susceptibility testing. All laboratory findings were cross-checked before entry into the study database to ensure accuracy and completeness.

Statistical Analysis

Data were entered, cleaned, and analyzed using appropriate statistical software. Descriptive statistics were employed for data analysis. Categorical variables were summarized as frequencies and percentages. The distribution of bacterial isolates and their antimicrobial susceptibility patterns were presented in tables. The findings were interpreted in relation to previously published literature.

Ethical Considerations

Ethical approval for the study was obtained from the appropriate institutional ethics committee prior to commencement of the research. Written informed consent was obtained from all participants or from parents/guardians in the case of minors. Confidentiality and anonymity of participants were maintained throughout the study, and all procedures were conducted in accordance with the ethical principles of medical research involving human participants.

RESULTS

Distribution of Gram-Negative Bacterial Isolates

Table I shows among the 83 bacterial isolates recovered from patients with chronic mucosal otitis media, 58 (69.9%) were Gram-negative organisms. Pseudomonas aeruginosa was the predominant pathogen, accounting for 36 (62.1%) of Gram-negative isolates, followed by Klebsiella pneumoniae 9 (15.5%), Escherichia coli 8 (13.8%), and Proteus species 5 (8.6%).

Table I

Distribution of Gram-negative bacterial isolates (*n*=58).

Organism	Frequency	Percentage (%)
Pseudomonas aeruginosa	36	62.1
Klebsiella pneumoniae	9	15.5
Escherichia coli	8	13.8
Proteus spp.	5	8.6

Antimicrobial Susceptibility Pattern of Pseudomonas aeruginosa

Table II presents the antimicrobial susceptibility profile of Pseudomonas aeruginosa. The highest susceptibility was observed for gentamicin (91.7%), followed by piperacillin-tazobactam (83.3%), meropenem (66.7%), and piperacillin (58.3%). High levels of resistance were noted against ampicillin (100%), ciprofloxacin (91.7%), amoxicillin-clavulanate (83.3%), ceftriaxone (75.0%), and aztreonam (75.0%).

Antimicrobial Susceptibility Pattern of Escherichia coli

The susceptibility pattern of Escherichia coli isolates is shown in *Table II*. The isolates demonstrated high sensitivity to amikacin (87.5%), meropenem (87.5%), and piperacillin-tazobactam (87.5%). Conversely, resistance was highest against ampicillin (87.5%), cefotaxime (87.5%), ceftriaxone (75.0%), and ciprofloxacin (62.5%).

Antimicrobial Susceptibility Pattern of Klebsiella pneumoniae

As illustrated in *Table II*, all Klebsiella pneumoniae isolates were susceptible to amikacin and meropenem (100% each). Piperacillin-tazobactam also showed good activity with 77.8% sensitivity. Complete resistance was observed against ampicillin, amoxicillin-clavulanate, and aztreonam.

Antimicrobial Susceptibility Pattern of Proteus Species
The antimicrobial susceptibility profile of Proteus species is presented in *Table II*. All isolates were susceptible to meropenem and piperacillin-tazobactam (100%), while 80.0% were susceptible to amikacin and ceftriaxone. Resistance was highest against ampicillin (100%), amoxicillin-clavulanate (80.0%), and aztreonam (80.0%).

Table II
Antimicrobial susceptibility pattern of Gram-negative bacterial isolates.

Antibiotic	E. coli (n=8) n (%)	P. aeruginosa (n=36) n (%)	K. pneumoniae (n=9) n (%)	Proteus spp. (n=5) n (%)
Ampicillin	1 (12.5)	0 (0.0)	0 (0.0)	0 (0.0)
Amoxicillin–clavulanate	3 (37.5)	6 (16.7)	0 (0.0)	1 (20.0)
Amikacin	7 (87.5)	21 (58.3)	9 (100.0)	4 (80.0)
Aztreonam	2 (25.0)	9 (25.0)	0 (0.0)	1 (20.0)
Cefotaxime	1 (12.5)	0 (0.0)	2 (22.2)	2 (40.0)
Ceftazidime	3 (37.5)	18 (50.0)	NT	NT
Co-trimoxazole	4 (50.0)	15 (41.7)	5 (55.6)	3 (60.0)
Ciprofloxacin	3 (37.5)	3 (8.3)	6 (66.7)	2 (40.0)
Ceftriaxone	2 (25.0)	9 (25.0)	1 (11.1)	4 (80.0)
Gentamicin	4 (50.0)	33 (91.7)	6 (66.7)	3 (60.0)
Meropenem	7 (87.5)	24 (66.7)	9 (100.0)	5 (100.0)
Piperacillin	NT	21 (58.3)	NT	NT
Piperacillin–tazobactam	7 (87.5)	30 (83.3)	7 (77.8)	5 (100.0)

NT = Not tested.

Overall Susceptibility Pattern of Gram-Negative Isolates
Overall, Gram-negative pathogens demonstrated substantial resistance to commonly prescribed antibiotics including ampicillin, amoxicillin-clavulanate, ciprofloxacin, and third-generation cephalosporins. Meropenem, amikacin, gentamicin, and piperacillin-tazobactam showed the highest antimicrobial activity against most Gram-negative isolates.

DISCUSSION

In this study, Gram-negative bacteria were the most prevalent among isolates from chronic mucosal otitis media, with Pseudomonas aeruginosa as the most frequently identified organism, followed by Klebsiella pneumoniae, Escherichia coli, and Proteus spp. Comparable results were observed in a tertiary care facility in Dhaka, Bangladesh, where Gram-negative bacteria were similarly identified as the primary pathogens, with Pseudomonas aeruginosa being the most common isolate [14]. The similarity among investigations emphasizes the persistent dominance of Gram-negative organisms, notably Pseudomonas aeruginosa, in chronic ear infections within the area.
In this study, Pseudomonas aeruginosa exhibited the greatest susceptibility to gentamicin and piperacillin–tazobactam, while demonstrating significant resistance to ciprofloxacin, ampicillin, and cephalosporins. E. coli, Klebsiella, and Proteus spp. showed the greatest sensitivity to meropenem, amikacin, and piperacillin–tazobactam, exhibiting significant resistance to β-lactam antibiotics. Comparable results were noted in a Bangladeshi tertiary care study, indicating a predominance of Gram-negative bacteria and increased susceptibility to aminoglycosides and carbapenems, coupled with notable resistance to frequently used antibiotics [15].
In general, Gram-negative bacteria—especially Pseudomonas aeruginosa—dominate in chronic mucosal otitis media with significant resistance to typical antibiotics, while aminoglycosides, carbapenems, and piperacillin–tazobactam continue to be the most effective, highlighting the necessity for culture-directed treatment.

CONCLUSION

The primary pathogens in chronic mucosal otitis media were Gram-negative bacteria, mainly Pseudomonas aeruginosa. The isolates demonstrated significant resistance to frequently used antibiotics like ampicillin, amoxicillin-clavulanate, and cephalosporins. In comparison, carbapenems, aminoglycosides, and piperacillin–tazobactam showed the greatest efficacy. The results emphasize the importance of continual monitoring of antimicrobial susceptibility trends and implementing culture-directed therapy to enhance treatment results and decrease antibiotic resistance.

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