Original research article

A Study of Microbiological Profile of Ear Infections with Antibiotic Sensitivity Pattern

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Abstract

Background: One of the most common diagnoses presented to ENT OPDs around the world is an ear infection. It is a significant health issue in developing nations like India. It has a connection to avoidable hearing loss. It carries a higher chance of developing potentially severe long-term consequences. Knowing their etiological agents as well as their antimicrobial sensitivity pattern will therefore aid in determining the best course of treatment and helping to avoid problems. The current research attempted to study the microbiological profile of ear infections with antibiotic sensitivity patterns.

Methods: Based on the inclusion and exclusion criteria a total of n=60 cases were identified during the period of study. The ear discharge was collected using sterile swabs under aseptic precautions. Three samples were taken. One swab for Direct Gram Stain and KOH mount. The Second swab was for bacterial culture and the third swab was for fungal culture. The pus swabs from discharge-producing ears were collected using swab methods and cotton wool. Stained with gram stain and followed by culture on MacConkey agar, Blood agar, Mannitol Salt agar, and Chocolate agar, ear discharge samples were inoculated.

Results: The commonest organism isolated as *P. aeruginosa* in 36.36% of cases followed by *S. aureus* in 25.45% of cases. *Proteus mirabilis, Acinetobacter baumannii, and Escherichia coli* were isolated in 5.45% cases each. Coagulase-negative *staphylococci* (CoNS), *K pneumoniae,* were isolated in 3.63% of cases. The fungal isolates that predominate include *Aspergillus niger 7.27% cases, followed by 1.81% of Aspergillus flavus, Candida tropicalis,* and *Candida* each.

Conclusion: According to this study, Staphylococcus aureus and Pseudomonas aeruginosa are the most typical causes of ear infections. The regularly prescribed medications Gentamycin, Amoxicillin, Erythromycin, and Cotrimoxazole have been proven to be less effective against these pathogens. Ciprofloxacin was responsive to the majority of the isolates.

Keywords: Ear Infections, Chronic Suppurative Otitis Media, Acute Otitis Media, Otitis Externa, Microbiological Profile.

Introduction

One of the most typical clinical presentations at ear, nose, and throat clinics in the tropics and subtropics is ear discharge. Due to low socioeconomic position, overcrowding, inadequate cleanliness, and malnutrition, ear infections are very common in India. otitis media is an inflammation of the middle ear cleft and the tympanum with otorrhoea lasting from two weeks to more than three months and a persistent perforation that is primarily brought on by bacteria. [1, 2] Ear infections can be either acute or persistently purulent. [3] Worldwide, there are between 65 and 330 million ear infections, and 60% of those persons have serious hearing loss. [4] Otitis media (OM) is a serious health issue that affects both industrialized and developing nations with a high incidence and prevalence. [5] Ear infections can affect adults, despite being largely a disease that affects newborns and young children. [6] The condition may start in childhood, develop as a side effect of untreated or improperly treated acute suppurative otitis media, or develop as a chronic condition right away. [5] A persistent perforation may provide the microbes access to the middle ear. Children are more likely to have ear infections than adults because the architecture of the eustachian tube makes it simpler for germs to enter the nasopharynx. Additionally, males are more likely to experience it than females. [5]

The majority of ear infections can be viral, bacterial, or fungal in origin. Ear infections have been linked to several bacterial species. Acute Otitis Externa is frequently associated with the isolation of *Pseudomonas aeruginosa*, *Streptococcus pyogenes*, and *Staphylococcus aureus*. Anaerobes and *pseudomonas* are frequently linked to chronic external otitis. *Streptococcus pneumoniae*, *Haemophilus influenzae*, and *Moraxella catarrhalis* are the most common pathogens in acute otitis media. *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Proteus* species, *Klebsiella* species, *Escherichia coli*, and anaerobes. and other bacteria are the main causes of chronic suppurative otitis media. [7] The introduction of more modern, broad-spectrum antibiotics has resulted in a continual change in the microbial flora. Group *A Streptococcus* was the most prevalent cause of acute otitis media in the pre-antibiotic period, but it is now rare. [8] The fungus may operate as the main pathogen or may be present in addition to bacterial infections. Humidity in the ear, an alkaline pH, epithelial debris and cerumen, immunocompromised status, and the use of topical steroids and antibiotics are among the factors that promote the growth of the fungus. A high prevalence of fungal infections is present in chronic otitis media patients as a result of the erratic and illogical use of antibiotics to treat bacterial infections. *Aspergillus* and *Candida* species are the two most prevalent fungi associated with ear infections. [9] Inappropriate use of antibiotics has decreased the frequency of problems while increasing the frequency of bacterial resistance. The majority of the time, empirical treatment is used, which has caused the emergence of numerous bacteria that are resistant to antibiotics. For the logical use of medications for therapy, current knowledge of the most frequent causing organism and their antimicrobial susceptibility pattern is crucial. In light of this, this study was conducted to identify the environmental causes of ear discharge, with a focus on the bacterial isolates' patterns of antibiotic susceptibility to facilitate effective treatment.

Material and methods

This cross-sectional study was conducted in the Department of Microbiology with the support of the Department of ENT, Prathima Institute of Medical Sciences, Nagunur, Karimnagar. Institutional Ethical approval was obtained for the study. Written consent was obtained from all the participants of the study.

Inclusion Criteria

1. Patients reporting ear discharge
2. Both male and female patients
3. Patients of all age groups

Exclusion Criteria

1. History of antimicrobial therapy in the last 7 days
2. Duplicate isolates of the same patient
3. Patients not willing to participate in the study voluntarily

Based on the inclusion and exclusion criteria a total of n=60 cases were identified during the period of study. The patient's History was obtained in terms of name, age, sex, duration of complaints and previous administration of antibiotics was noted in a predesigned proforma. The ear discharge was collected using sterile swabs under aseptic precautions. Three samples were taken. One swab for Direct Gram Stain and KOH mount.

The Second swab was for bacterial culture and the third swab was for fungal culture. The pus swabs from discharge-producing ears were collected using swab methods and cotton wool. [1] Stained with gram stain and followed by culture on MacConkey agar, Blood agar, Mannitol Salt agar, and Chocolate agar, ear discharge samples were inoculated. In a candle jar that can produce about 5% CO_2, Blood and Chocolate agar plates were incubated. The inoculated media were all incubated for 18 –24 hours at 37 °C. Manual identification of bacterial species was done using standard microbiological techniques. Potassium hydroxide (KOH) wet mount: A drop of 10% KOH was placed on a clean glass slide containing the specimen to digest the keratin surrounding the fungi. A clean coverslip was placed over it without air bubble formation and examined under 10X and 40X objectives after 20 minutes for the presence of fungal elements like budding yeast cells, hyphae, and spores. The isolates were subjected to antimicrobial susceptibility testing by a modified Kirby-Bauer disc diffusion method and interpreted as per CLSI 2018 guidelines. [10]

***Statistic evaluation*:**

SPSS statistical software was used to enter and analyze the data (IBM Corp., 2011). New York: IBM Corp., IBM SPSS Statistics for Windows, Version 20.0. Percentages were used to express discrete variables. P values of 0.05 were regarded as statistically significant when comparing the proportion of bacterial isolates with the patient's age and sex using the chi-square test.

Results

Based on clinical diagnosis of the cases included in the study n=3 were cases of otitis externa, n=10 were acute otitis media, n=40 were cases of chronic suppurative otitis media, and n=7 were cases of otomycosis. A critical analysis of table 1 shows that most of the cases included in the study were aged 11 – 20 years with 28.33% of cases, followed by 21 – 30 years with 18.33% of cases. Out of n=60 cases, n=25(42%) were females and n=35(58%) were males depicted (Figure 1). The male to female ratio was 1.4: 1. The mean age of the cases in the study was 19.56 ± 8.5 years.

Table 1: Age Distribution of cases included in the study

Age Group	*No. of Cases*	*Percentage*

01 - 10	6	10.0
11 - 20	17	28.33
21 - 30	11	18.33
31 - 40	9	15.0
41 - 50	8	13.33
51 - 60	6	10.0
61 - 70	2	3.33
71 - 80	1	1.67
Total	60	100

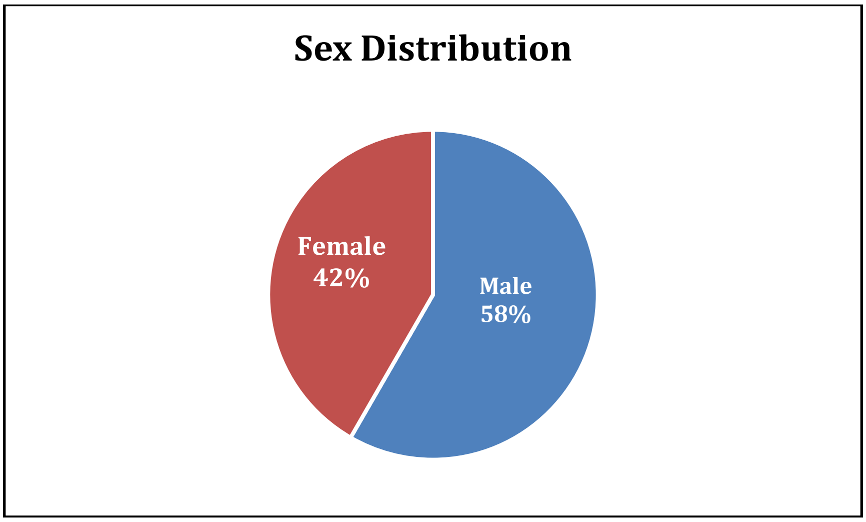

Figure 1: Sex-wise distribution of the cases included in the study

Out of n=60 cases, n=56 was unilateral 93.33% and n=4 (6.67%) was with the involvement of bilateral ears. Right-sided ear involvement was seen in n=37 cases and left side in n=19 cases the percentage-wise distribution has been depicted in figure 2.

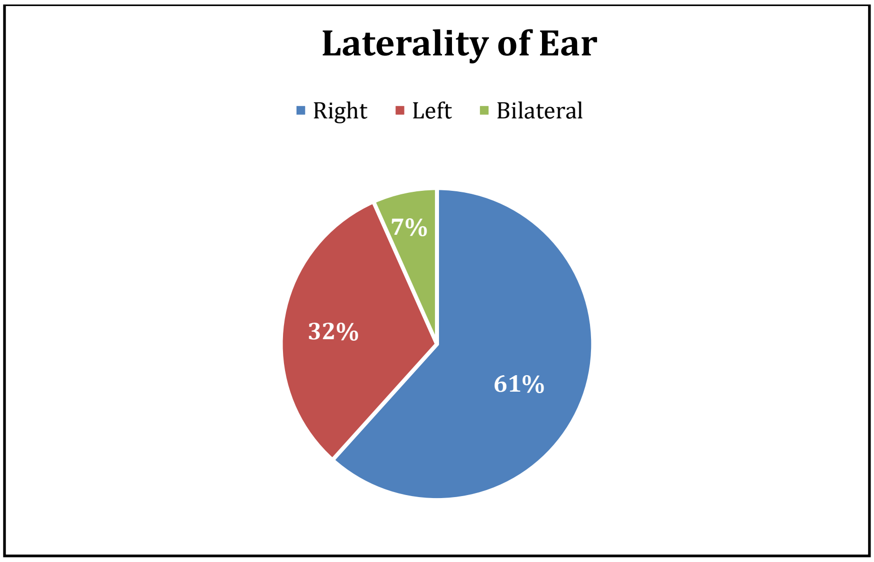

Figure 2: Laterality of ear involved in the cases of the study

Out of n=60 samples obtained and sent for the culture, we found n=55(91.67%) cases were culture positive and n=5(8.33%) cases were culture negative. Among the culture-positive cases, 58.18% were gram-negative bacilli and 29.09% were gram-positive cocci and 12.72% were fungal isolates details depicted in table 2.

Table 2: Microbiological profile of isolates from the cases

Isolates	No. of Cases	Percentage
Gram-positive cocci	16	29.09
Gram Negative Bacilli	32	58.18
Fungi	7	12.72
Total	55	100.0

The commonest organism isolated was *P. aeruginosa* in 36.36% of cases followed by *S. aureus* in 25.45% of cases. *Proteus mirabilis, Acinetobacter baumannii, and Escherichia coli* were isolated in 5.45% cases each. Coagulase-negative *staphylococci* (CoNS), *K pneumoniae*, were isolated in 3.63% of cases. The fungal isolates that predominate include *Aspergillus niger 7.27% cases, followed by 1.81% of Aspergillus flavus, Candida tropicalis,* and *Candida* each. The details of the isolation of the bacterial and fungal isolates have been depicted in table 3.

Table 3: Culture Isolates from the positive cases of the study

Organism	No. of isolates	Percentage
Pseudomonas aeruginosa	20	36.36
Staphylococcus aureus	14	25.45
CoNS	2	3.63
Klebsiella pneumoniae	2	3.63
Proteus mirabilis	3	5.45
Acinetobacter baumannii	3	5.45
Proteus vulgaris	1	1.81
Escherichia coli	3	5.45
Aspergillus niger	4	7.27
Aspergillus flavus	1	1.81
Candida tropicalis	1	1.81
Candida glabrata	1	1.81
Total	55	100

Based on the antibiotic sensitivity pattern out of the n=20 *Pseudomonas aeruginosa* isolates, maximum resistance was noted for ceftazidime (65.00%), followed by Gentamycin resistance (35.00%). The sensitivity of the organisms to Ciprofloxacin, Amikacin, and Piperacillin Tazobactam was 80.00%, 85.00%, and 90.00% respectively all the n=20 *Pseudomonas aeruginosa* isolates showed 100% sensitivity to Meropenem all the n=20 *Pseudomonas aeruginosa* isolates showed 100% sensitivity to Meropenem i.e., no Metallo-beta-lactamases (MBL) was detected depicted in table 4.

Table 4: Antibiotic Sensitivity Testing of *Pseudomonas aeruginosa (n=20)*

Drug	*Sensitive (%)*	*Resistant (%)*
Ceftazidime	45.00	65.00
Gentamycin	65.00	35.00
Ciprofloxacin	80.00	20.00
Amikacin	85.00	15.00
Piperacillin-Tazobactam	90.00	10.00
Meropenem	100.00	00.00

Out of the n=14 *Staphylococcus aureus* isolates, maximum resistance i.e., 78.57% was noted for Penicillin, followed by Cotrimoxazole 57.14% and Erythromycin 42.85%.

Table 5: Antibiotic Susceptibility Testing of *Staphylococcus aureus (n=14)*

Drug	*Sensitive %*	*Resistant %*
Penicillin	21.03	78.57
Cotrimoxazole	42.85	57.14
Erythromycin	57.14	42.85
Clindamycin	85.71	14.28
Ciprofloxacin	85.71	14.28
Cefoxitin	92.85	07.14
Vancomycin	100.00	00.00

Among the n=14 *Staphylococcus aureus* isolates n=2 cases were resistant to Cefoxitin were MRSA isolates (14.28%) given in table 6.

Table 6: MRSA isolates in *Staphylococcus aureus*

MRSA	*Frequency*	*Percentage*
Positive	2	14.28
Negative	12	85.71
Total	14	100

Discussion

The most common condition requiring patients to see a doctor and take medication is an ear infection. [11] One of the often-requested specimens for culture and antimicrobial susceptibility tests from clinical settings in the research area is an ear discharge sample. A total of n=60 patients with symptoms of ear infection attending the ENT department were included in the study. Male and female patients in all age groups from 1 to 80 years were included the mean age of the cases in the study was 19.56 ± 8.5 years. The age distribution analysis in our study shows peak incidence in the second decade of life (28.33%) the mean age of the cases in the study was 19.56 ± 8.5 years. A similar analysis was made by Derese Hailu et al., [12] which showed that 63% of cases occurred in 0 to 20 years of age. This could be brought on by inadequate care, poor hygiene, poverty, and crowding. [13] Another typical risk factor for young adults is an upper respiratory infection. Gulathi et al., [14] found that the fifth decade of life had the highest prevalence (27%) followed by the third decade (25.42%) and the first decade. Due to their undeveloped immune systems and short, broad, and more horizontal Eustachian tubes, children may have a higher prevalence of upper respiratory tract infections. Out of n=60 cases, n=25(42%) were females and n=35(58%) were males and the male to female ratio was 1.4: 1. Madana et al., [15] reported a similar observation, finding that men (56 percent) were more affected than women (44%). There is no information on the distribution of ear infections by gender, however, male preponderance may be related to their more exposed, active lifestyles or the manner they clean their ears. The right ear was commonly involved in 61% of cases and 32% were with left ear involvement and bilateral involvement was noted in 7% of cases of the study. Right side predominance was because most persons are right-handed, the right ear was afflicted more frequently than the left which can be attributed to recurrent infections there. Arun Ghosh et al., [16] in a similar study found cases with 3.70 percent of bilateral cases, 66.66 percent right-sided, and 29.62 percent left-sided ear infections, in agreement with observations of the current study. The culture positivity rate of this study was 91.67% and culture negative was 8.33%. No growth in culture may be due to prior antibiotic usage or infection by strict anaerobes or viral agents. [17] Among the culture-positive cases, most of the organisms were aerobic bacterial isolates and 12.27% were fungi. This is consistent with research by Srivastava A et al., [18] which found that bacteria are more prevalent than fungi (77% versus 23%). Fungal development is caused by the fact that cerumen and environmental fungal spores settle in the external auditory canal's wetness and warmth. [19] Opportunistic fungi may arise as a result of prolonged topical, broad-spectrum antibiotic instillation that suppresses the natural bacterial flora. Among the Gram-Positive cocci (29.09%), the most common was *Staphylococcus aureus* (25.45%), followed by Coagulase Negative *Staphylococcus* (3.63%). This is in concordance with the study results of Raghu Kumar *et al.,* [20] *Staphylococcus aureus* 34.44% and Coagulase Negative *Staphylococcus* 3.33%. The most common cause of otomycosis in the current study was *Aspergillus niger* which can be attributed to the fact that they can grow even in nutrient-depleted conditions. The current study demonstrates that Pseudomonas aeruginosa and Staphylococcus aureus are the most frequent causes of ear infections in our system. Ciprofloxacin, Amikacin, and Piperacillin Tazobactam are bactericidal medications that can be taken safely by people of all ages and are extremely effective against these isolates. A semi-synthetic ureidopenicillin is a piperacillin. Due to its wide spectrum of Penicillin and anti-pseudomonal action, it is effective against both Gram Positive and Gram-Negative bacteria. A beta-lactamase inhibitor, tazobactam binds to the active site of the enzyme to block beta-lactamase activity, restoring the activity of primary beta-lactam antibiotics. In our investigation, Piperacillin-Tazobactam was sensitive to 90.0% of Pseudomonas aeruginosa. Amikacin is a semisynthetic aminoglycoside antibiotic with the widest spectrum. In our study, 85% of *Pseudomonas aeruginosa* were sensitive to Amikacin. In *Pseudomonas aeruginosa* cases no Metallo-beta-lactamases (MBL) were detected since in our OPD most ear infections are treated with amoxicillin and erythromycin, therefore, the sensitivity to organisms to antibiotics like amikacin and piperacillin-tazobactam was higher and we have found any case of Metallo- β-Lactamase producing *Pseudomonas aeruginosa.* However, if these antibiotics are used improperly, resistant bacteria may develop. Thus, it is crucial to use these antibiotics wisely and to abide by the antibiotic policies. To give the correct antibiotic, in the right dose, at the right time, requires regular evaluation and understanding of the microbiological profile and their antimicrobial sensitivity pattern in that particular region. In addition to preventing the establishment and spread of resistant strains, this will aid in avoiding consequences that could develop if the infection was not appropriately treated.

Conclusion

Ear infections are very common presentation and early detection of the etiological agents and knowledge of their antibiotic sensitivity pattern can help reduce the occurrence of ear infections. According to this study, Staphylococcus aureus and Pseudomonas aeruginosa are the most typical causes of ear infections. The regularly prescribed medications Gentamycin, Amoxicillin, Erythromycin, and Cotrimoxazole have been proven to be less effective against these pathogens. Ciprofloxacin was responsive to the majority of the isolates. Various geographic regions have different strains of bacteria that cause ear infections, and antibiotic resistance pattern the occurrence of resistant bacterial species, the resistance profile also differs proportionally. Therefore, testing for antimicrobial susceptibility should be performed on all isolates to assist in selecting the right antibiotics for therapy.

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