PSEUDOMONAS AERUGINOSA IN CHRONIC SUPPURATIVE OTITIS MEDIA: SENSITIVITY SPECTRUM AGAINST VARIOUS ANTIBIOTICS IN KARACHI

Tahira Mansoor, Mohammed Ayub Musani*, Gulnaz Khalid, Mustafa Kamal**

Department of Biotechnology, University of Karachi, Karachi, Pakistan. *Department of ENT, **Department of Pathology, Karachi Medical and Dental College and Abbasi Shaheed Hospital, Karachi, Pakistan

Background: Chronic suppurative otitis media (CSOM) is a prevailing and notorious infection in developing countries causing serious local damage and threatening complications. Early and effective treatment based on the knowledge of causing micro organisms and their sensitivity results in good clinical recovery and prevents from damage and complications. *Pseudomonas aeruginosa* is most common pathogen causing CSOM in Pakistan. The objective of this study is to identify incidence of *Pseudomonas aeruginosa* involved in CSOM and sensitivity against commonly prescribed antibiotics. **Methods:** A total of 263 patients with unilateral or bilateral chronic suppurative otitis media (CSOM) were enrolled in the study at Department of ENT, Karachi Medical and Dental College & Abbasi Shaheed Hospital, Karachi, Pakistan from December 2004 to May 2006. Sterile cotton swabs were used to collect pus from discharging ears and plated on Blood agar and MacConkey agar for 24 to 48 hours. Antibiotics susceptibility was tested by Minimum inhibitory concentrations (MIC) method using Mueller-Hinton agar. **Results:** Overall microbiology of 267 samples from 263 patients was studied including 4 of bilateral discharge. Polymicrobial growth was present in 8 samples. A total of 275 bacterial isolates were studied. *Pseudomonas aeruginosa* (40%) and *Staphylococcus aureus* (30.9%) were the most common bacterial agents found in CSOM. MIC was done for *Pseudomonas aeruginosa* only as it was the commonest pathogen found in CSOM. Sensitivity pattern of *Pseudomonas aeruginosa* showed that amikacin was active against 96% of isolates followed by ceftazidime 89%, ciprofloxacin 85%, gentamicin 81%, imipenem 76%, aztreonam 42% and ceftriaxone 21%. **Conclusions:** *Pseudomonas aeruginosa* was the most common bacteria isolated from chronic discharging ears followed by *Staphylococcus aureus.* Amikacin was found to be the most suitable drug followed by ceftazidime and ciprofloxacin for *Pseudomonas aeruginosa*. The resistance against ceftriaxone and aztreonam was found to be very high.

Keywords: Chronic suppurative otitis media, Antibiotic resistance, *Pseudomonas aeruginosa*

INTRODUCTION

Otitis media is infection of middle ear caused by bacteria, fungi and virus resulting in inflammation of mucosal lining. Recurrent Otitis media may cause damage of ossicles, facial nerve and cochlea, resulting in permanent hearing loss.[1] It can be acute or chronic. The acute form usually associated with the infection in the upper respiratory tract whereas persistent form is known as chronic suppurative otitis media (CSOM).[2]

The chronic form is still a major problem in developing countries like Pakistan. It is more common in children belonging to lower socioeconomic group. Most common micro organisms found in CSOM are *Pseudomonas aeruginosa, Staphylococcus aureus, Proteus mirabilis, Klebsiella pneumoniae, Escherichia coli, Aspergillus spp* and *Candida spp* but these organisms vary in various geographical areas.[1]

Antimicrobial therapy is used to eradicate the bacterial agents causing otitis media but most of the micro organisms are acquiring antibiotic resistance. In developing countries, this problem is rapidly increasing due to misuse of antibiotics. The important factors associated with its occurrence were found to be poor hospital hygiene, overcrowding, lack of resources for infection control, and a lack of personnel trained in controlling infections in hospital.[3]

Antibiotic resistance can be natural or acquired.[4] There are certain mechanism for the causes of bacterial resistance[5] such as:

a) The presence of enzymes that inactivates the antimicrobial agent(s).
b) Mutation in the antimicrobial agents target(s), which reduces the binding of the antimicrobial agent.
c) Reduced uptake of the antimicrobial agent(s).
d) Overproduction of target of antimicrobial agent(s).

This study was undertaken to identify frequency of *Pseudomonas aeruginosa* involved in patients of CSOM and the sensitivity pattern to commonly prescribed antibiotic in our local community.

MATERIAL AND METHODS

This study was carried out at the out patient department of Karachi Medical & Dental College and Abbasi Shaheed Hospital, Karachi. Some clinical isolates from discharging ear were also collected from other laboratories of Karachi.

A total of 263 patients with unilateral or bilateral CSOM were enrolled after detail clinical

history regarding the age, duration of ear discharge and especially any antibiotic treatment received. Though clinical examination was done to rule out acute otitis media and otitis externa. Sterile cotton swabs were used to collect pus from discharging ears. Patients were of both genders and from all age groups. Inclusion and exclusion criteria were set as follows:

Patient of any age, patient of any sex, ear discharge of more than 3 months duration were included. Discharge of less than 3 months duration, discharge with intact tympanic membrane (otitis externa) and patient receiving antibiotics at presentation were excluded.

Overall microbiology of 275 samples was studied during 18 months from December 2004 to May 2006. The swabs were plated on MacConkey agar (Oxoid CM115), Blood agar (Oxoid CM55) and Chocolate agar and incubated for 24 to 48 hours at 37°C. Identification of bacterial strains was done by colonial morphology on selective and differential medium. All isolates were biochemically tested for TSI, SIM and citrate test in the laboratory from where the organisms were collected.

One hundred and ten (110) isolated strains of *Pseudomonas aeruginosa* were tested for susceptibility to antibiotics by agar dilution method using Mueller-Hinton agar (Oxoid CM337). The antibiotics tested were amikacin (Bristol Myers Squibb), gentamicin (Reckitt Benckiser), ciprofloxacin (Sami Pharmaceuticals), ceftazidime (Glaxo Wellcome), ceftriaxone (Bosch Pharmaceuticals), imipenem (Merck Sharp & Dohme) and aztreonam (Bristol Myers Squibb). Minimal inhibitory concentration (MIC) of these antibiotics were monitored according to the guideline of National Committee for Clinical Laboratory Standards.[6]

RESULTS

A total of 263 patients were selected for the study among them 4 had bilateral discharges. All 267 samples showed single while polymicrobial growth was present in 8 samples. A total of 275 bacterial isolates were studied during 18 months. Age of patients ranges from 6 months to 70 years. Children and young adults were more affected and together comprise about 71% of total cases (Figure-1). Females (52%) were more commonly affected than males (48%).

Pseudomonas aeruginosa (40%) was the most common bacterial agent found in chronic discharging ear, followed by *Staphylococcus aureus* (30.9%), *Proteus* (11.6%), *Klebsiella* (8%) and *E. coli* (4%) (Table-1).

Antibiotic susceptibility was tested by Minimum inhibitory concentration (MIC) method using Mueller-Hinton agar. Sensitivity pattern of *Pseudomonas aeruginosa* showed that amikacin was active against 96% of isolates followed by ceftazidime 89%, ciprofloxacin 85%, gentamicin 81%, imipenem 76%, aztreonam 42% and ceftriaxone 21% (Figure-2).

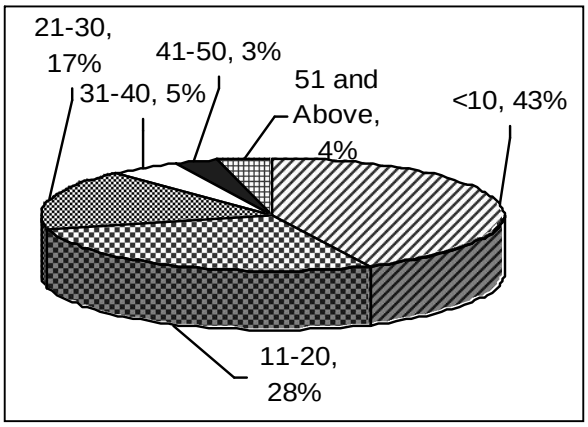


Figure-1: Age distribution

Table-1: Microbiological profile of bacteria in CSOM

Type of organisms	No. of isolates (n=275)	%
1. Gram (+) bacteria		
Staphylococcus aureus	85	30.9
Staphylococcus epidermidis	10	3.6
Streptococcus spp.	05	1.8
2. Gram (-) bacteria		
Pseudomonas aeruginosa	110	40
Proteus mirabilis	32	11.6
Klebsiella spp.	22	8
Escherichia coli	11	4

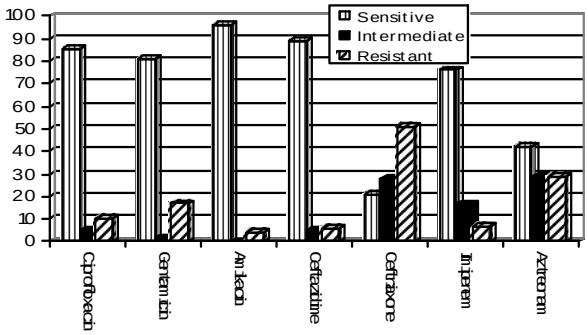
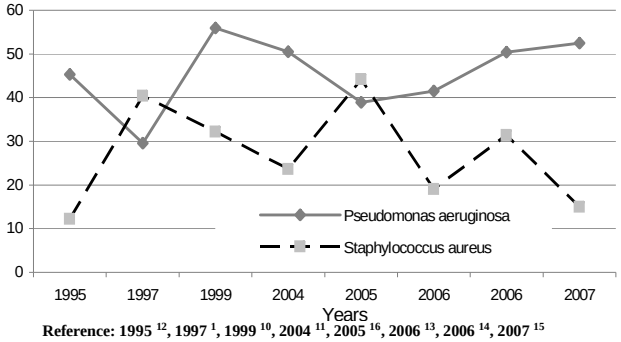


Figure-2: Antibiotic profile of *Pseudomonas aeruginosa*



Reference: 1995 [12], 1997 [1], 1999 [10], 2004 [11], 2005 [16], 2006 [13], 2006 [14], 2007 [15]

Figure-3: Comparison of occurrence of *Pseudomonas aeruginosa* & *Staphylococcus aureus* in CSOM from various studies in Pakistan

DISCUSSION

Chronic suppurative otitis media (CSOM) and various complications associated with the disease such as irreversible local destruction of middle ear structures, facial palsy, serious intracranial and extracranial complications are among the most common conditions seen by the otologist, paediatrician and the general practitioner. It is a persistent disease and often causes irreversible local destruction of middle ear.[2]

In our study, CSOM was found mostly among children and in young adults (Figure-1). Same results were obtained in Nigeria and India.[7,8] This may be due to multiple reasons as young children and infants have low resistance and also because of relative short and straight Eustachian tube (inner ear tube). However, in Singapore, 23.3% cases belonged to the age group of 31–40 years.[9]

Females were more commonly affected than males and it is also supported by a study carried out in Singapore.[9] In contrast to this some studies showed opposite trend reported male 57.3% and female 42.7%[10] and this can be due to geographical variation.

Our results show that CSOM in Pakistan is mainly due to *Pseudomonas aeruginosa* followed by *Staphylococcus aureus*. However, *Proteus mirabilis*, *Klebsiella*, *E. coli* etc was also found (Table-1).

In some studies carried out in different cities of Pakistan, the results of their bacterial isolates were similar in order of frequency to the results obtained in our study[10–15] but some showed different trend[1,16] also shown in Figure-3 and this could be due to variation in microorganisms in different locality and effect of climate.[17]

As *Pseudomonas aeruginosa* was the most prevalent microorganism that is also supported by national and international studies[7,9,18,19] antibiotics resistant profile was done against seven antibiotics. These antibiotics belong to the group of aminoglycosides (Amikacin, gentamicin), flouroquinolones (ciprofloxacin), cephalosporins (ceftazidime, ceftriaxone), carbapenem (imipenem), and monobactam (aztreonam).

Aminoglycosides are bactericidal antibiotics that interfere with protein synthesis and most frequently used because of its activity against gram negative bacteria. We found that *Pseudomonas aeruginosa* were sensitive to aminoglycosides, i.e., amikacin and gentamicin and it is also supported by previous studies in Nigeria[7], Nepal[18], India[20] and in Pakistan as well.[10,11,13,15] In contrast, some studies showed that *Pseudomonas* acquiring resistant against gentamicin.[14,19]

Flouroquinolones have a broad range of activity and it is found to be active against *Pseudomonas aeruginosa*. It inhibits the bacterial DNA gyrase or the topoisomerase II, thereby inhibiting DNA replication and transcription. The resistance to flouroquinolones is basically a reflection to mutation which is a result of selective pressure created by the use of it.[21] A study conducted in South Korea, reported that in adult patients with chronic suppurative otitis media (CSOM), ciprofloxacin resistant *Pseudomonas aeruginosa* was found.[2] In our study, 85% of *Pseudomonas aeruginosa* was found to be sensitive to ciprofloxacin and in various studies conducted in Pakistan, it is also reported that more than 90% of isolates were sensitive to ciprofloxacins.[1,10,11,14,15] It shows that ciprofloxacin is very effective in our community.

Cephalosporins are the most frequently prescribed class of antibiotics and third-generation display an extended gram negative spectrum. These drugs are also used in treating pseudomonal infections. If *Pseudomonas aeruginosa* have become resistant to one cephalosporin are often resistant to other β-lactam anti-pseudomonas agents as well as to other antibiotics.[22] It is indicated that ceftazidime is very effective to treat nosocomial infections and pediatric patients[23,24] but not active against *Pseudomonas aeruginosa* that causing CSOM and other otogenic complications.[2,25] Some studies showed that ceftriaxone is also a drug of choice and can be used in combination of other antibiotics.[25] In Pakistan, it was found that third generation cephalosporins were very effective against bacterial isolates found in CSOM.[13,14,15] In present study, 89% of isolates were found to be sensitive to ceftazidime and it is also comparable to other studies.[10] On the other hand, only 21% of isolates showed sensitivity to ceftriaxone in our findings. These results suggested that in our local community, ceftazidime is effective but resistance against ceftriaxone increases progressively.

Imipenem belongs to carbapenem group of antibacterial and it binds to different penicillin binding proteins. It is the most active drug against *Pseudomonas aeruginosa* found in CSOM.[2,14,15] In our study, 76% of isolates were found to be sensitive while 17% showed intermediate pattern and 7% were found to be resistant. Imipenem is very effective against *Pseudomonas aeruginosa* but may be prolonged treatment of *Pseudomonas aeruginosa* infected patients with this antibiotic has often allowed the emergence of imipenem resistant mutants.[26]

Similarly, aztreonam is a monobactam antibiotic with virtually no gram positive activity but good gram negative activity. It is also active against β-lactamase producing organisms. Some local studies showed that *Pseudomonas aeruginosa* was sensitive to aztreonam[1,15] but in present study more than 50% of isolates were found to be resistant. This is an agreement to the observation of Somekh *et al* that

aztreonam is an optional alternative systemic treatment for patients with pseudomonal CSOM.[27]

Pseudomonas can grow well in the absence of special nutrition, it proliferates at room temperature, and it is highly resistant to antibiotic, making it difficult to treat. Antibiotic therapy for the treatment of CSOM is preferable, depending upon kind of causative agent and patient's age. However, the lack of consistent availability of necessary antibiotics limits the therapeutic choice. The main cause of CSOM is failure of treatment of acute otitis media. On the other hand the use of expired or degraded antibiotics, the prescription of illogical antimicrobial combination by unskilled physicians and practitioners and in many cases the lack of sufficient amount supplied because of individual economical constrains has also played a role in the development of antimicrobial resistance. So, it is very much needed that the doctors should have knowledge about the changing pattern of causative agents against drugs and should not prescribe such antibiotics and in addition to that patients also should be educated to avoid misuse of antibiotics.

CONCLUSIONS

Pseudomonas aeruginosa was the most common pathogen followed by *Staphylococcus aureus* isolated from CSOM.

Amikacin was found to be the most suitable antibiotic followed by ceftazidime and ciprofloxacin for *Pseudomonas aeruginosa*. The resistance against ceftriaxone and aztreonam was found to be very high. The important factor that is responsible for resistance is inappropriate duration and dose of antibiotics. Most of the commonly used antibiotics become ineffective to *Pseudomonas aeruginosa*. It is essential to use antibiotics in proper way to prevent emergence and spread of resistant pathogens.

REFERENCES

1. Anwar-us-Salam, Abid SH, Abdulla EM. Suppurative Otitis in Karachi: An Audit of 510 Cases. Pak J Otolaryn 1997;13:66–9.
2. Jang CH, Park SY. Emergence of ciprofloxacin-resistant Pseudomonas in chronic suppurative otitis media. Clin Otolaryngol 2004;29:321–3.
3. Hart CA, Kariuki S. Antimicrobial resistance in developing countries. BMJ 1998;317:647–50.
4. Normark BH, Normark S. Evolution and spread of antibiotic resistance. J Intern Med 2002;252:91–106.
5. Fluit AC, Visser MR, Schmitz FJ. Molecular Detection of Antimicrobial Resistance. Clin Microbiol Rev 2001;14:836–71.
6. National Committee for Clinical Laboratory Standards. Performance Standards for Antimicrobial Susceptibility Testing. NCCLS, Wayne, PA, USA 2001.M100–S10.
7. Wariso BA, Ibe SN. Bacteriology of chronic discharging ears in Port Harcourt, Nigeria. West Afr J Med 2006;25:219–22.
8. Poorey VK and Lyer A. Study of bacterial flora in CSOM and its clinical significance. Indian J Otolaryngol Head Neck Surg 2002;54:91–5.
9. Loy AHC, Tan AL, Lu PKS. Microbiology of chronic suppurative otitis media in Singapore. Singapore Med J 2002;43:296–9.
10. 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.
11. 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.
12. Ahmad M, Amjad M, Hameed A. Microflora in chronic suppurative otitis media. Specialist. 1995;12:19–22.
13. Iqbal SM, Udaipurwala IH, Hasan A, Shafiq M, Mughal S. Chronic suppurative otitis media: Disease pattern and drug sensitivity. J Surg Pak 2006;11:17–9.
14. Ahmed S, Iqbal MA, Hassan Z, Khurshid T, Ali L, Pervez Q. Spectrum and bacterial isolates in chronic suppurative otitis media in Khuzdar. Pak J Otolaryngol 2006;22:34–6.
15. Gul AA, Ali L, Rahim E, Ahmed S. Chronic suppurative otitis media; frequency of Pseudomonas aeruginosa in patients and its sensitivity to various antibiotics. Professional Med J 2007;14:411–5.
16. Ahmed B, Hydri AS, Afridi, AAK, Ejaz A, Farooq S, Zaidi SK. Microbiology of ear discharge in Quetta. J Coll Physicians Surg Pak 2005;15:583–4.
17. Yildirim A, Erdem H, Kilic S, Yetiser S, Pasha A. Effect of climate on the bacteriology of chronic suppurative otitis media. Ann Otol Rhinol Laryngol 2005;114:652–5.
18. Sharma S, Rehan HS, Goyal A, Jha AK, Upadhyaya S and Mishra SC. Bacteriological profile in chronic suppurative otitis media in Eastern Nepal. Trop Doct 2004;34:102–4.
19. Yeo SG, Park DC, Hong SM, Cha CI, Kim MG. Bacteriology of chronic suppurative otitis media-a multicentre study. Acta Otolaryngol 2007;127:1062–67.
20. Saini S, Gupta N, Aparna, Seema, Sachdeva OP. Bacteriological study of pediatric and adult chronic suppurative otitis media. Indian J Pathol Microbiol 2005;48:413–6.
21. Sheng WH, Chen YC, Wang JT, Chang SC, Luh KT, Hsieh WC. Emerging fluoroquinolone-resistance for common clinically important gram negative bacteria in Taiwan. Diagn Microbiol Infect Dis 2002;43:141–7.
22. Carsenti-Etesse H, Cavallo JD, Roger PM, Ziha-Zarifi I, Plesiat P, Garrabe E *et al*. Effect of beta-lactam antibiotics on the in vitro development of resistance in Pseudomonas aeruginosa. Clin Microbiol Infect 2001;7:144–51.
23. Sevillano E, Valderrey C, Canduela MJ, Umaran A, Calvo F, Gallego L. Resistance to antibiotics in clinical isolates of Pseudomonas aeruginosa. Pathol Biol 2006;54:493–7.
24. Jones RN, Sader HS, Fritsche TR, Pottumarthy S. Comparisons of parental broad-spectrum cephalosporins tested against bacterial isolates from pediatric patients: report from the SENTRY Antimicrobial Surveillance Program (1998–2004). Diag Microbiol Infect Dis 2007;57:109–16.
25. Hafidh MA, Keogh I, Walsh RM, Walsh M, Rawluk Daniel. Otogenic intracranial complication. A 7-year retrospective review. Am J Otolayngol 2006;27:390–5.
26. Okamoto K, Gotoh N, Nishino T. Pseudomonas aeruginosa reveals high intrinsic resistance to penem antibiotics: penem resistance mechanisms and their interplay. Antimicrob Agents Chemother 2001;45:1964–71.
27. Somekh E, Cordova Z. Ceftazidime versus aztreonam in the treatment of pseudomonal chronic suppurative otitis media in children. Scand J Infect Dis 2000;32:197–9.

Address for Correspondence:

Dr. Mustafa Kamal, Associate Professor, Department of Biotechnology, University of Karachi, Karachi-75270, Pakistan. **Cell:** +92-333-3613509

Email: mustafacamal@hotmail.com