Original Article

Bacterial isolates and drug susceptibility patterns of ear discharge from patients with ear infection at Shaheed Monsur Ali Medical College.

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Abstract

Ear infection is a common problem for both children and adults especially in developing countries. However in Bangladesh particularly in the study area, there is no recent data that shows the magnitude of the problem. A retrospective study was conducted from from April, 2013 to March, 2014 at Shaheed Monsur Ali Medical College. Ear discharge samples were cultured on MacConkey agar, blood agar and chocolate agar plates. A total of 115 ear discharge samples were tested for bacterial isolation and 86 (74.78%) cases were found positive; 49 (56.98%) were gram negative bacteria and the predominant isolate were *S. aureus* 21 (24.42%) followed by *Pseudomonas aeruginosa* 20 (23.26%). Of individuals 70 (81.40%) had single bacterial infection and 16 (18.60%) had mixed bacterial infections. Under five children were more affected by ear infection. The prevalence of ear infection was higher in females than males (64.35% vs 35.65%) (P=0.879). The prevalence of ear infection was very high in the study area. Majority of the bacterial isolates were resistant to multiple antibiotics. Hence antibiotics susceptibility test is mandatory before prescribing any antibiotics.

Key words: Antibiotic susceptibility, Bacterial isolates, Bangladesh, Ear infection.

INTRODUCTION

Ear infection is a common problem for both children and adults especially in developing countries. Ear infection is an inflammation of the ear and ear discharge is one of the commonest symptoms of ear infection.¹ About 65-330 million people suffer from ear infection worldwide and 60% of them had significant hearing impairment.² Otitis media is an inflammation of the middle ear that affects the tympanic membrane.³ Clinically, otitis media may be classified as acute, sub acute, and chronic suppurative otitis media (CSOM).⁴ CSOM is a chronic suppurative inflammation of the middle ear cleft leading to dangerous complications like meningitis and subdural abscess in the absence of timely management. Though ear infection can be caused by viruses and fungi, the major causes of ear infections are bacteria such

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as Pseudomonas aeruginosa, Staphylococcus aureus, Proteus mirabilis, Klebsiella pneumoniae and Escherichia coli which are found in the skin of the external ear and enter into the middle ear through a chronic perforation.^{5,6} In addition, antimicrobial resistance profile of bacteria varies among population because of the difference in geography, local antimicrobial prescribing practices and prevalence of resistant bacterial strains in a given area.⁷ So there should be up to date information on microbial resistance pattern at national and local levels to guide the rational use of the existing antimicrobial drugs. Since the introduction of antibiotics, complications have become less common. However, their occurrence should not be underestimated due to the morbidity and mortality associated with them. Microbial culture and sensitivity helps in appropriate management of CSOM.⁴ The purpose of this study was to acquire data on the pattern of CSOM caused by aerobic bacteria and to determine the antibiotic sensitivity of the isolated organisms prevalent in our hospital.

MATERIAL AND METHODS

A retrospective study was conducted from April, 2013 to March, 2014 at Shaheed Monsur Ali Medical College and

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Hospital, Bangladesh.

Study participants and data collection

Samples were collected from 115 clinically diagnosed cases of CSOM by ENT specialists and processed according to standard protocols. Patients with symptoms of CSOM and who were not on antibiotics were included in the present study. Socio-demographic and laboratory results which contain different bacterial isolates and drug susceptibility patterns of patients were collected from the Hospital Microbiology Laboratory unit registration books by using standard data collection format.

Culture and identification

According to the standard operation procedures, the ear discharge samples were collected aseptically by using cotton swab techniques from different OPDs and wards of the hospital and transported to microbiology laboratory. Ear discharge samples were cultured on MacConkey agar, blood agar and chocolate agar plates and then incubated aerobically at 37°C for 24 hours. Pure isolates of bacterial pathogen were preliminary characterized by colony morphology, gram-stain and catalase test. Bacterial species were identified as per the standard microbiological methods.⁸

Antimicrobial susceptibility testing

Antimicrobial susceptibility tests were done on Mueller-Hinton agar (Oxoid, England) using disk diffusion method.⁹ Gram positive and gram-negative bacterial sensitivity of isolates to commonly used antimicrobials (gentamicin, ciprofloxacin, amoxicillin, ceftriaxone, cefixime, cephradine, cefuroxime, doxycycline, chloramphenical, erythromycin, cotrimazole and levofloxacin) were investigated (Oxoid, England). The drug susceptibility pattern was interpreted according to Clinical and Laboratory Standards Institute (CLSI, 2012)¹⁰ Reference strains of *E. coli* ATCC 25922 and *S. aureus* ATCC 25923 were used for quality control for antimicrobial susceptibility tests.¹⁰

RESULTS

A total of 115 ear discharge samples were analyzed at the Medical College Hospital Microbiology Laboratory unit during the study period. Majority of the study participants were females (64.35% vs 35.65%) (Table 1). The mean age of the study participants was (19 ± 12) years with the minimum and maximum age of 2 years and 75 years respectively. The overall prevalence of bacterial isolation was 86 (74.78%). From the total bacterial isolates, 49 (56.98%) were gram negative bacteria. Of individuals who were culture positive, 70 (81.40%) had single bacterial infection while 16 (18.60%) had mixed bacterial infections. In this study, the predominant bacterial isolates were *S. aureus* 21 (24.42%) followed by

Table 1: Result of culture from ear discharge in different sex.

Sex	Culture Positive n (%)	Negative n (%)	_Total n (%)				
Female	55 (63.95)	19 (65.52)	74 (64.35)				
Male	31 (36.05)	10 (34.48)	41 (35.65)				
Total	86 (100)	29 (100)	115 (100)				
$X^2 = 0.231$, df = 1, P = 0.879.							

Pseudomonas aeruginosa 20 (23.26.5%). Other predominant bacterial isolates includes coagulase negative *Staphylococcus species*, *E. coli*, *Proteus* and *Klebsilla species*. Majority (22.09%) of the bacterial isolates were found in under five age groups (Table 2).

 Table 2 : The distribution of bacterial isolates from ear discharge in age catagories of study participants.

Age in		Bact	erial	Isolates	No	o (%)	
	S.aureus	E. coli	Pseud.spp	Kleb.spp	Prot.spps	CNStaph spps	Total
0-5	9(47.37)	9(47.37)	6(31.58)	0	0	1(5.26)	19(22.09)
6-10	2(13.33)	2(13.33)	2(13.33)	1(6.67)	2(13.33)	7(46.67)	15(17.44)
11-15	3(30)	3(30)	1(10)	2(20)	2(20)	1(10)	10(11.63)
16-20	2(16.67)	2(16.67)	2(16.67)	3(25)	2(16.67)	2(16.67)	12(13.96)
21-30	1(10)	1(10)	0	1(10)	1(10)	3(30)	10(11.63)
31-40	2(40)	2(40)	1(20)	0	1(20)	1(20)	5(5.81)
>40	2(13.33)	2(13.33)	8(53.33)	1(6.67)	1(6.67)	1(6.67)	15(17.44)
Total	21(24.42)	21(24.42)	20(23.26)	8(9.30)	9(10.47)	16(18.60)	86(100)
$\overline{X^2} = 4$	45.7, df	= 30, P=	= 0.033.				

Among the *S. aureus*, 71.43% were resistant to erythromycin and 80% of the *Pseudomonas species* were resistant to chloramphenicol, erythromycin and cefixime. About 85% *S. aureus* and *Pseudomonas* species were sensitive to levofloxacine. Of these, 83.33% *E. coli* were resistant to cephradine, 75% *Klebsiella* were resistant to amoxaicillin and 81.25% coagulase negative Staphylococcus species were resistant if cefixime (Table 3). From the total bacterial isolates, 74 (86.05%) had multiple antibiotic resistant pattern (resistant to two or more antibiotics) and 9 (10.47%) of the isolates were resistant to at least one antibiotic. Only 3 (3.49%) bacterial isolates were susceptible to all antibiotics. Bacteria isolated fram ear discharge with antibiotic susceptibility

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Table 3: Antimicrobial resistance pattern of bacterial isolates

 from ear discharge samples of study participants.

	Resistant pattern of bacterial isolates, n(%)					
Antimicrobial agents	S. aureus (n=21)	E.coli 1 (n=12)	Pseud.spps (n=20)	Kleb. spp (n=8)	Prot.spps (n=9)	CNstap.spps (n=16)
AMX	8(38.10)	8(66.67)	15(75)	6(75)	7(77.78)	3(18.75)
CFM	9(42.86)	6(50)	16(80)	5(62.50)	4(44.44)	13(81.25)
CN	3(14.29)	4(33.33)	8(40)	2(25)	2(22.22)	3(18.75)
CRO	3(14.29)	2(16.67)	5(25)	1(12.5)	1(11.11)	2(12.5)
CIP	7(33.33)	3(25)	5(25)	1(12.5)	3(33.33)	4(25)
CE	6(28.57)	10(83.33)	15(75)	4(50)	3(33.33)	7(43.75)
С	4(19.05)	3(25)	16(80)	3(37.50)	4(44.44)	5(31.25)
CXM	5(23.81)	3(25)	14(70)	5(62.50)	4(44.44)	4(25)
DO	3(14.29)	6(50)	15(75)	4(50)	4(44.44)	7(43.75)
Е	15(71.43)	5(41.67)	16(80)	2(25)	6(66.67)	10(62.50)
SXT	10(47.62)	5(41.67)	17(70)	3(37.50)	4(44.44)	8(50)
LEV	3(14.29)	4(33.33)	3(15)	1(12.50)	1(11.11)	1(6.25)

AMX= Amoxicillin, CFM= Cefixime, CN= Gentamycin, CRO= Ceftriaxone, CIP= Ciprofloxacin, CE= Cephradine, C= Chloramphenicol, CXM= Cefuroxime, DO= Doxycycline, E= Erythromycin, SXT= Cotrimoxazole, LEV= Levofloxacin.

DISCUSSION

In this study, 74.78% cases of ear discharges were found to be positive for bacteria, which is in agreement with other studies in India 11 and Iran 12. Monomicrobial growth was seen in 81.40% of cases, which is similar to the previous study by Agarwal et al.13 In our study, majority of the patients were found less than 20 years of age, which was also documented in previous studies.^{14, 15} In contrast, Lov et al showed the increased prevalence in 30-40 years age in his study.¹⁶ In our study, 64.35% were females and 35.65% were males. Thus females were affected more in our study which is in accordance with Loy et al 16, but differ from data of Ahmed et al ¹⁷. Females were more affected by ear infections. This may be due to the difference between ear cleaning habit of the males and females. In some tradition, females use cotton swabs to clean their ear and this may contribute for the introduction of microorganisms from the external surface to the middle ear. However in some studies^{18,19} there were no difference on the prevalence of ear infections between males and females. The result of this study showed that Staphylococcus aureus (24.42%) was the commonest isolates followed by Pseudomonas species (23.26%) which is similar to previous study.^{11, 12} This may be due to the difference in climate and geographical variations in different countries. Coagulase negative Staphylococci were the next common

organisms. It is usually considered as commensal organism of the skin, but rarely becomes an opportunistic pathogen. The other organisms which have been isolated in the present study in descending order were E. coli (13.95%), Proteus (10.47%) and Klebsiella species (9.30%) which were also isolated in previous study.^{11, 16} In the present study, different bacterial species had different level of resistance pattern to different antibiotics. However, Staphylococcus aureus were sensitive to commonly used antimicrobials and showed high sensitivity rates to chloramphenicol (80.71%), gentamicin and doxycycline (85.71%). Clinical resistance of Staphylococci spp. to penicillin and other antimicrobial agents is now a problem throughout the world.²⁰⁻²² Pseudomonas species showed high sensitivity to levofloxacin (88.89%) and relatively sensitive to ciprofloxacin and cefixime (77.78%). Although resistant strains of Pseudomonas isolates to fluoroquinolones were detected in other studies.²³⁻²⁵ In this study, 81.25% of coagulase negative Staphylococci were resistant to cefixime, 75% of Klebsiella species and 77.78% of Proteus were resistant to amoxicillin. Similar findings were also reported in other studies.12 Prescription of antibiotics without laboratory guidance and over sales of antibiotics without proper drug prescription may be some of the different factors that can contribute for this high level drug resistant pattern. Therefore, drug prescription for patients should be laboratory evidence based.

Conclusion

In conclusion, the overall prevalence of bacterial isolates was high and majority of the isolates were gram negative bacteria. The predominant isolates were *S. aureus* and Pseudomonas species. The bacteria which have been isolated from otitis media have shown high level of antibiotic resistance in the study area. Majority of the bacterial isolates were resistant to multiple antibiotics.

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