

Original Research Articles

Bacterial isolates from Otitis Media infections and their antibiograms, Hodeidah city, Yemen

ABSTRACT:

Aims: This study was aimed to determine the predominant bacterial isolates and their antimicrobial susceptibility patterns from ear discharge among children and adults in Yemen.

Study design: Original research article.

Place and Duration of Study: Department of Medicine and Department of Microbiology, Al-Thawrah and Al-Amal hospitals, Hodeidah city, Yemen, between August 2014 to March 2015.

Methodology: Ear swabs collected from 80 patients with otitis media were inoculated into blood, chocolate, MacConkey agar plates and subjected to several bacteriological tests and analysis. Isolated bacterial pathogens were tested against thirteen antibiotics using standard bacteriologic techniques.

Results: The most common isolated bacteria were Gram positive (27=42.9%) of *Staphylococcus aureus* and Gram negative (21=33.3%) of *Pseudomonas aeruginosa*. Gentamycin, Tetracyclin, Ciprofloxacin, and Norfloxacin revealed high level of sensitivity to *Staphylococcus aureus*, whereas Polymyxin-B and Cotrimoxazol to *Pseudomonas aeruginosa*.

Conclusion: *Staphylococcus aureus* and *Pseudomonas aeruginosa* were the predominant pathogens of discharging ear. In the study area, gentamicin and tetracycline would be the drug of choice in *Staphylococcus aureus* isolates, while *Pseudomonas aeruginosa* was highly sensitive to polymyxin-B and cotrimoxazol.

Key words: Otitis media, bacterial pathogens, resistance pattern

1. INTRODUCTION

Otitis media (OM) is the inflammation of the middle ear, which may be caused by bacteria, fungi or viruses. There are three types of otitis media, and these are, acute purulent otitis media, otitis media with effusion and chronic suppurative otitis media [1,2].

Otitis media is a major health problem of children in developing countries with poor socioeconomic status. This is due to the fact that children are more prone to otitis media related to the immaturity of their immune status, the shorter and horizontal nature of Eustachian tubes, frequent exposure to upper respiratory tract infections and malnutrition [3-6].

The significant risk factors in OM include socioeconomic status, cultural, seasonal, and age factors, as well as family history of the middle ear disease. Other risk factors include: environmental tobacco smoke, crowded living conditions, daycare attendance, pacifier use, and bottle feeding in a supine position. Hand washing, breastfeeding, and immunizations offer some protection against otitis media [7-14].

In developing countries, chronic suppurative otitis media (CSOM) occurs as a complication of acute otitis media with perforation and can be a major health issue. The range of bacterial pathogens associated with CSOM is considerably broader than those seen in AOM. *Pseudomonas*, *Staphylococcus*, *Proteus*, and *Klebsiella* species are most commonly isolated, and mixed infections are common [15-18].

Acute otitis media (AOM) is the most common indication for the use of antimicrobial agents in children [19-22].

The aims of this study were to isolate and identify the bacterial etiology causing Otitis Media, and to determine their susceptibility pattern to the most common used antibiotics.

2. METHODOLOGY

2.1 Study Population:

This cross-sectional prospective study was carried out during the period of August 2014 to March 2015. A total of 80 samples from patients (37 male and 43 female) with otitis media infection were seen at the ENT clinics at Al-Thawrah hospital and Al-Amal hospital, Hodeidah city, Yemen; and that were clinically diagnosed to have otitis media. An informed consent was obtained from each patient to collect an ear discharge by the attending physician and filled a predesigned questionnaire, which included questions regarding age, sex, living place, symptoms, medications, previous OM infection(s), and family history of OM.

2.2 Sample collection, handling, culture and identification:

Ear swap/discharge specimens were aseptically collected. Patient's ear was washed by normal saline (0.85% NaCl). Discharges samples were collected using sterile cotton swabs. All ear swabs/discharge were directly inoculated onto blood, chocolate, MacConkey agar (Company, Country). The Blood and MacConkey agar plates were incubated aerobically, while chocolate agar plates were incubated anaerobically under 5% CO₂ atmosphere both at 37°C for 24-48 hours. All positive cultures were identified according to standard microbiology methods, including: cultural characters, Gram stain, biochemical reactions such as: coagulase, catalase, citrate media and Kilgler Iron Agar (KIA), urease test, indole test, oxidase and diagnostic tests such as gas production, bacitracin and optichin.

2.3 Antimicrobial susceptibility testing:

Antimicrobial susceptibility tests were performed for all isolates according to the criteria of the (CLSI) clinical and laboratory standards institute. Bacterial suspension was prepared and was adjusted to a McFarland solution 0.5 and inoculated onto Mueller-Hinton agar (Company. Country) using disk diffusion method. The susceptibility pattern of the bacterial pathogens was determined towards the following antimicrobial agents, tetracycline, polymyxine-B, erythromycin, gentamicin, ciprofloxacin, norfloxacin, cotrimoxazole, ceftriaxone, amoxicillin, cefotaxime, cefuroxime, chloramphenicol, and clarithromycin.

2.4 Statistical analysis:

Statistical analysis was done using SPSS software version 15. Chi-square test was employed to compare the proportion of bacterial isolates with patients' age and comparison of antimicrobial resistances. P-value of < 0.05 was considered to indicate statistically significant difference.

3. RESULTS

Table (1) shows the distribution of Otitis Media infection according to demographic characteristics of the studied sample. It was evident that the highest infection was 23(36.5%) at age <10 years, followed by the age category 20-30 years 15(23.8%). Approximately the infections were divided between males and females. Majority of infected sample was collected from urban patients 49 (77.8%). Most of the infected patients were illiterates, which showed highly statistically significant relationship (<0.001).

Table 1. Distribution of Otitis Media infection according to demographic characteristics

Demographic Characteristics		Positive(63)		Negative(17)		Total(80)		X ²	p-value
		No	%	No	%	No	%		
Age	<10	23	36.5	7	41.2	30	37.5	72.5	0.13
	10-	14	22.2	3	17.6	17	21.3		
	20-	15	23.8	6	35.3	21	26.3		
	30-	3	4.8	1	5.9	4	5.0		
	40-	5	7.9	0	0	5	6.3		
	50-	2	3.2	0	0	2	2.5		
	60-70	1	1.6	0	0	1	1.3		
Gender	Total	63	100%	17	100%	80	100%	7.3	0.50
	Male	30	47.6	7	41.2	37	46.3		
	Female	33	52.4	10	58.8	43	53.8		
	Total	63	100%	17	100%	80	100%		
Residence	Urban	49	77.8	12	70.6	61	76.3	5.2	0.74
	Rural	14	22.2	5	29.4	19	23.8		
	Total	63	100%	17	100%	80	100%		

Education	University	11	17.5	5	29.4	16	20	95.7	0.00
	Secondary	9	14.3	3	17.6	12	15		
	Preparatory	3	4.8	0	0	3	3.8		
	Primary	3	4.8	0	0	3	3.8		
	Professional	1	1.6	0	0	1	1.3		
	Illiterate	36	57	9	53	45	56.3		
Antibiotic Use	Total	63	100%	17	100%	80	100%	15.3	0.05
	Yes	24	38.1	5	29.4	29	36.3		
	No	39	61.9	12	70.6	51	63.8		
	Total	63	100%	17	100%	80	100%		
Collection site	Left Ear	24	38.1	9	52.9	33	41.3	9.9	0.27
	Right Ear	39	61.9	8	47.1	47	58.8		
	Total	63	100%	17	100%	80	100%		

Table (2) demonstrates the distribution of isolated bacteria from patients with Otitis Media according to gender. It was noticed that the most common bacterial infection caused by *Staphylococcus aureus* bacteria (42.9%) followed by *Pseudomonas aeruginosa* (33.3%). Males were infected more than females by *Staphylococcus aureus*, while females were infected more by *Pseudomonas aeruginosa*. From the isolated bacteria, females were totally infected more than males.

Table 2. Distribution of isolated bacteria from patients with Otitis Media according to gender

Bacteria	Male	%	Female	%	Total	%
<i>Staphylococcus aureus</i>	16	53.3	11	17.4	27	42.9
<i>Pseudomonas aeruginosa</i>	8	26.7	13	20.6	21	33.3
<i>Proteus vulgaris</i>	1	3.3	3	4.7	4	6.3
<i>Proteus mirabilis</i>	2	6.7	2	3.2	4	6.3
<i>Klebsiella pneumonia</i>	1	1.6	2	3.2	3	4.8
<i>Escherichia coli</i>	1	1.6	0	0.0	1	1.6
<i>Providencia species</i>	0	0	1	1.6	1	1.6
<i>Salmonella paratyphi</i>	0	0	1	1.6	1	1.6
<i>Other Salmonellae</i>	1	1.6	0	0.0	1	1.6

Total	30	47.7	33	52.3	63	100
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Table (3) illustrates the distribution of isolated bacteria from patients with Otitis Media. It was clear that Gram negative bacteria were more than Gram positive (57.1% to 42.9%). *Staphylococcus aureus* was the only Gram positive bacteria isolated (42.9%), while the most common Gram negative bacteria was *Pseudomons aeruginosa* (33.3%).

Table 3. Distribution of isolated bacteria from patients with Otitis Media

Gram stain	Bacteria	Number	%
Gram positive	<i>Staphylococcus aureus</i>	27	42.9
	<i>Pseudomons aeruginosa</i>	21	33.3
	<i>Proteus vulgaris</i>	4	6.3
	<i>Proteus mirabilis</i>	4	6.3
Gram negative	<i>Klebsiella pneumonia</i>	3	4.8
	<i>Escherichia coli</i>	1	1.6
	<i>Providencia species</i>	1	1.6
	<i>Salmonella paratyphi</i>	1	1.6
	<i>Other Salmonellae</i>	1	1.6
	Total	63	100

Table (4) shows the distribution of isolated bacteria from Otitis Media patients according to antimicrobial drugs sensitivity. It was found that the most abundant isolated bacteria was *Staphylococcus aureus* (27(43%)) which was sensitive mainly to both Gentamicin and Tetracycline (89% each) followed by both Norfloxacin and Ciprofloxacin (81% each). *Pseudomons aeruginosa* was the second abundant isolated bacteria (21(33%)) and was mainly sensitive to Polymyxin-B and Cotrimoxazol (100% and 86% respectively). Cotrimoxazol, Gentamicin, Ciprofloxacin, and Norfloxacin were the most antimicrobial drugs showed an effect on different isolated bacteria.

Table 4. Distribution of isolated bacteria from Otitis Media patients according to sensitivity to antimicrobial drugs

Isolated Bacteria	Sensitivity to antimicrobial drugs											
	Tetracycline	Erythromycin	Chloramphenicol	Gentamicin	Ciprofloxacin	Norfloxacin	Cotrimoxazol	Ceftriaxone	Amoxacilin	Cefotaxime	Clarithromycin	Polymyxin-B
<i>Staphylococcus aureus</i> (27)	24	1	1	24	22	22	19	3	17	7	5	1
<i>Pseudomons aeruginosa</i> (21)	2	3	4	9	10	9	18	1	2	3	4	21

<i>Proteus vulgaris</i> (4)	1	--	1	2	3	3	3	3	1	1	2	1	--	
<i>Proteus mirabilis</i> (4)	3	--	--	3	3	3	--	3	--	1	2	--	--	
<i>Klebsiella pneumonia</i> (3)	3	--	1	3	2	3	3	--	--	1	1	--	1	
<i>Escherichia coli</i> (1)	--	--	--	--	--	--	--	--	--	--	--	1	--	
<i>Salmonella paratyphi</i> (1)	--	--	--	--	--	1	--	--	--	--	--	--	--	
<i>Other Salmonellae</i> (1)	--	--	--	--	1	--	--	1	1	--	--	--	--	
<i>Providencia species</i> (1)	1	--	1	1	1	1	1	1	--	--	--	--	--	
Total (63)	No.	34	4	8	42	42	42	44	12	21	13	13	11	23
	%	54	6	13	67	67	67	70	19	33	21	21	17	37

4. DISCUSSION

The term “otitis media” covers a wide spectrum of disease, and is used to describe illnesses with predominantly middle ear symptoms (including acute otitis media, otitis media with effusion, and chronic suppurative otitis media [23,24].

In the study area, otitis media is common health problem in children more than adults. Among children, peak-age prevalence was observed in the age group under ten years (<10). This is comparable with previous studies conducted in Thamar [25], Ethiopia [26,27] and Nigeria [28].

Similar to a study was done in Pakistan [29]; the present study indicated that OM is more prevalent in females (53.75%) than males (46.25%). In contrast, the study in Thamar and Nigeria reported that males were affected more than females [25,28].

The present study is comparable to those reported in Ethiopia [26] and Iraq [30], where generally the Gram negative bacteria were dominant isolates (57.1%) of discharging ears compared to Gram positive bacteria (42.9%).

Staphylococcus aureus and *Pseudomonas aeruginosa* were the most dominant isolates, in agreement with studies conducted in Thamar [25], Ethiopia [26] and Basrah [30], and oppose to other studies in India and Nigeria, where *Pseudomonas aeruginosa* was isolated more than *Staphylococcus aureus* [28,31].

The most similar study to this study was that done in Iran in which the most isolated pathogen was *Staphylococcus aureus* followed by *Pseudomonas aeruginosa*, and *Proteus species* [32].

In this study *Staphylococcus aureus* revealed a high level of sensitivity to gentamicin, ciprofloxacin, norfloxacin and cotrimoxazol, while a study in Ethiopia showed high level of resistance to amoxicillin, and low resistance to ciprofloxacin, gentamicin and norfloxacin [26]. *Pseudomonas aeruginosa* was the most antibiotic resistance microorganism isolated in otitis media, most of the isolates were resistant to amoxicillin, erythromycin, tetracyclin, and chloramphenicol, which is comparable to a study in Ethiopia [26], however, *Pseudomonas aeruginosa* was highly sensitive to polymixin-B and cotrimoxazol, in contrast to that study in Ethiopia which showed high sensitivity to norfloxacin, ciprofloxacin and gentamicin [26].

5. CONCLUSION

Otitis media was a common health problem of all age groups especially children. *Staphylococcus aureus* and *Pseudomons aeruginosa* were the predominant pathogens of discharging ear. In the study area, gentamicin and tetracycline (89% each) would be the drug of choice in *Staphylococcus aureus* isolates, while *Pseudomons aeruginosa* was highly sensitive to polymyxin-B and cotrimoxazol (100% and 86% respectively).

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