

Research Article

Estimation of the Antibiotic Activity against *Pseudomonas* spp Isolated from Ear Infection

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A B S T R A C T

Background: An ear infection, can be classified into otitis externs and otitis media, this affects all age groups especially infants and young. This infection associate with pathogenic microorganism type, frequent antibiotic uses, health care and age. The most common pathogenic bacteria of the ear infection are *Pseudomonas* spp. Antibiotic resistance represents a serious threat to the health of humans.

Methods: 48 ear swabs were collected through the use of wooden sticks in a sterile container for the identification of *Pseudomonas*, *Serratia* and *Klebsiella* species by VITEK 2 system and biochemical tests. The antibiotic susceptibility against these bacterial species was detected through the method of standard disk diffusion on the Moller Hinton agar.

Results: Among ear swabs, the positive growth percentage of the pathogenic gram-negative bacteria was 29.166%, and the percentage of *Pseudomonas* spp. was 57.142%. Males were found to be more susceptible than females with an infection percentage of 57.142%.

Conclusion: The antibiotic susceptibility patterns show the azithromycin, gentamycin, piperacillin, ceftriaxone, cefepime, imipenem, ceftriazone, and gentamycin have activity against *Pseudomonas* spp.

Keywords: Ear Infection, *Pseudomonas*, *Serratia*, *Klebsiella*, Antibiotics

Introduction

The infection of the ear is a common disorder for both children and adults, and this differs from one country to the other. Ear infections can be classified into otitis media and otitis externs. It represents the major health of the public, especially in the developing countries which associate with high disease burden and the economic impact to the patients.¹ Otitis media is inflammation due to bacterial infection of a middle ear cleft, tympanum and lasting from two weeks into more than three months.^{2,3} Ear infections can be acute or chronic.⁴ However, about 56 to 330 million

people suffer from ear infections worldwide, and about 60% of them suffer from loss of significant hearing.⁵ This infection can cause impaired speech, poor development of school language and social interactions.⁶ Although ear infections can affect people of all ages, but infants and young are more susceptible, due to the presence of a short Eustachian tube which eases the entry of pathogenic microorganisms to the nasopharynx.⁷ The preponderance of the microbiological investigations had explained that the most pathogenic bacteria associated with ear infections are *Pseudomonas* and *Klebsiella*,⁸ and the major causative agent associated with an ear infection in addition to

Pseudomonas aeruginosa is *Klebsiella* spp.⁹ In addition, the *Serratia marcescens* is associated with infections of otitis media.¹⁰ In spite of the natural protection mechanisms that have antibacterial effects, opportunistic pathogens such as *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, and other pathogenic bacteria can enter through a pierced ear.¹¹ Moreover, the signs and symptoms of ear infections differ depending on the pathogenic microorganism, such as bacteria or virus and/ or fungi, which leads to the use of antibiotics irrespective of the causative agents. This leads to big economic loss, stress to ear patients, and also increases the rates of antibiotic resistance.¹² As per the World Health Organization (WHO), the resistance to antibiotics represents one of the major global problems and a threat to human health.¹³

Material and Methods

Samples

The samples of 48 ear swabs were collected through the use of a sterile wooden stick. These samples were placed in a sterile container and transferred to the laboratory at the Imam Ali hospital in the Babylon Province, Iraq. Afterwards, they were used for the isolation and identification of *Pseudomonas*, other gram-negative bacteria and for studying the antibiotic susceptibility against these bacterial species.

Identification of the Gram-Negative Bacteria

The gram-negative bacteria were included species of the *Pseudomonas*, *Serratia* and *Klebsiella* which collected from ear smear swabs. These bacterial species were identified through the use of the VITEK 2 system and biochemical tests, and this is explained in Table 1.

Table 1. Identification of Pathogenic Gram-Negative Samples

Pathogenic bacteria	Procedure
<i>Pseudomonas</i> spp	VITEK 2 System & biochemical tests
<i>Serratia</i> spp	
<i>Klebsiella</i> spp	

Detection of the Antibiotics Susceptibility

Detection of the antibiotic susceptibility against *Pseudomonas*, *Serratia* and *Klebsiella*, which isolated from ear swabs were carried out by the method of standard disk diffusion on the medium of Moller Hinton ager by using several types of antibiotics. This procedure involved the incubation of these bacterial species together with used antibiotics on the Moller Hinton medium for 24h. Then after this period measured the inhibition zones formed in bacterial species cultures by antibiotics using a special millimetre ruler.

Results

Determine the type of pathogenic bacteria associated with ear infections and antibiotics, which use in treating this infection type represent the goal of a large number of researchers. The samples of ear swabs have been collected for isolation and diagnosis of *Pseudomonas* and *Serratia* species and study the antibiotics susceptibility against these bacterial species. Table 2 shows the age category of 5-14 years showed more bacterial growth (42.857%). It also reveals that men were more susceptible than women (infection percentages - 57.142% and 42.857% respectively). Table 3 explains the percentage of samples with bacterial growth were 29.166% among all ear swabs. Table 4 shows the *Pseudomonas* spp were more pathogenic bacteria isolated among pathogenic gram-negative bacteria (57.142%). Table 5 explains that azithromycin, gentamycin, piperacillin, ceftriaxone, cefepime, imipenem, ceftriaxon and gentamycin have activity against *Pseudomonas* spp., trimethoprim, amikacin, imipenem and levofloxacin have activity against *Serratia* spp. and levofloxacin and amikacin have activity against *Klebsiella* spp.

Table 2. Prevalence of Pathogenic Gram-Negative Bacteria according to Age Categories and Gender

Age Categories (years)	Positive No (%)	Negative No (%)
5 - 14	6 (42.857)	10 (29.411)
15 - 24	2 (14.285)	5 (14.705)
25 - 34	4 (28.571)	12 (35.294)
35 - 44	2 (14.285)	7 (20.588)
Gender		
Male	8 (57.142)	23 (67.647)
Female	6 (42.857)	11 (32.352)

Table 3. Comparison between the Samples with Bacterial Growth, Positive and Negative Bacteria

Bacterial Growth	Percentage
Positive	29.166
Negative	70.833

Table 4. Comparison between the Species of Pathogenic Gram-Negative Bacteria in the Samples with Bacterial Growth

Bacterial Species	Percentage
<i>Pseudomonas</i> spp	57.142
<i>Serratia</i> spp	28.571
<i>Klebsiella</i> spp	14.285

Table 5. Antibiotic Susceptibility against Pathogenic Gram-Negative Bacteria

Pathogenic bacterial	Antibiotic sensitive	Antibiotic resistant
<i>Pseudomonas</i> spp	Azithromycin Gentamycin Piperacillin Ceftriaxone Cefepime Imipenem Ceftriazon Gentamycin	Ceftriaxone Nalidixic acid Ciprofloxacin Rifampicin Trimethoprim/Sulfonamide Trimethoprim Amoxiclav
<i>Serratia</i> spp	Trimethoprim Amikacin Imipenem/Levofloxacin	
<i>Klebsiella</i> spp	Levofloxacin Amikacin	Nalidixic acid Ciprofloxacin Methoprim Amoxicillin

Discussion

In the present study, the younger ages (0-14 years) were more susceptible to ear infections. In a study by Addas F et al., who found the majority of ear patients were of the young ages (0-17 years).¹⁴ The even though infection of the ear is primarily a disorder of young children, but can affect adults.¹⁵ The high frequency of ear infections among young ages, may be due to short of the board and straight nature of the Eustachian tube, lower immunity, frequent infection of upper respiratory tract, malnutrition and lack of hygiene.¹⁶⁻¹⁸ In the current study, males were more susceptible than females. According to Hailu D et al., ear infections were more common in males than females (92.7% and 65% respectively).¹⁹ While Addas F et al. detected that this infection was higher in females than men with 65.8% and 34.2%, respectively.¹⁴ Argaw-Denboba A et al. demonstrated both males and females were at equal risk by 50.4% and 49.1%, respectively.²⁰ The few differences in the number of males and females having ear infections in the present study are unclear, but it may be due to variations in behaviour, care-seeking and study area.²¹ In this study, the positive growth percentage among ear swab samples was 29.166%. In a study by Hailu D et al., it was found that the positive percentage of gram-negative bacteria was 58.8% of ear infection isolates.¹⁹ Anie CO et al. demonstrated that the percentage of gram-negative bacteria positive was 23.9% among this infection.²² In the present study, the percentage of *Pseudomonas* spp, *Serratia* spp, and *Klebsiella* spp of the ear swab isolate was equal to 57.142%, 28.571%, and 14.285%, respectively. A study by Kittinger C et al. showed that the percentage of *Pseudomonas* spp of the ear infection is 66.0%.²³ Whereas Uddén F et al. demonstrated that the percentage of *Pseudomonas* and *Klebsiella* spp is

13.2% and 3% respectively.²⁴ Also, Appiah-Korang L et al. identified that the percentage of *Pseudomonas* spp, *Serratia* spp and *Klebsiella* spp is 46%, 2.2% and 5.6% respectively, among ear swabs.²⁵ Wasihun AG, Zemene Y, who found the percentage of *Pseudomonas* spp and *Klebsiella* spp to be 16.7% and 11.1% respectively.⁴ In the current study, azithromycin, gentamycin, piperacillin, ceftriaxone, cefepime, imipenem, ceftriazon and gentamycin show activity against *Pseudomonas* spp, while trimethoprim, amikacin, imipenem and levofloxacin shows activity against *Serratia* spp. Levofloxacin and amikacin show activity against *Klebsiella* spp. Appiah-Korang L et al. found that the antibiotic sensitivity percentage against *Pseudomonas* spp, was amoxicillin (8.1%), TMP-SMX (92%), ampicillin (28.5%), ceftriaxone (22.7%), ciprofloxacin (90.3%) and gentamicin (75.5%); while against *Klebsiella* spp, which was amoxicillin (12.5%), TMP-SMX (27.8%), ampicillin (0%), ceftriaxone (23.5%), ciprofloxacin (100%) and gentamicin (79.3%).²⁵ While Gorems K et al. demonstrated the resistance percentage of antibiotic against *Pseudomonas aeruginosa* which was ceftriaxone (100%), ciprofloxacin (21.4%), cefepime (64.3%), gentamicin (14.3%) and amikacin (21.4%); whereas against *Klebsiella* spp., which was ampicillin/ amoxicillin (10%), amoxicillin clavulanic acid (62.5%), ceftriaxone (37.5%), ciprofloxacin (0%), cefepime (25%), gentamicin (37.5%), sulfamethoxazole-trimethoprim (62.5%) and cefuroxime (62.5%).¹ Also Hailu D et al. detected the resistance percentage of antibiotics against *Pseudomonas aeruginosa* which was ampicillin (90%), amoxicillin/clavulanic acid (89%), ceftriaxone (23%), amikacin (4.5%), gentamycin (10.2%), ceftazidime (6.8%) and piperacillin (10.2%); while against *Klebsiella pneumoniae* which was ampicillin (100%), amoxicillin/

clavulanic acid (89%), ceftriaxone (20%), amikacin (0%), gentamycin (20%), ceftazidime (20%) and piperacillin (10%).¹⁹ Wasihun AG, Zemene Y, found that the resistance percentage of antibiotic against *Pseudomonas aeruginosa* was norfloxacin (80%), amoxicillin clavulanic acid (93%), trimethoprim-sulphamethoxazole (80%), penicillin (93%), ampicillin (86.7%), ceftriaxone (60%), ciprofloxacin (20%) and gentamicin (20%); whereas against *Klebsiella* spp which was norfloxacin (50%), amoxicillin-clavulanic acid (72.2%), trimethoprim-sulphamethoxazole (77.8%), penicillin (93%), ampicillin (86.7%), ceftriaxone (60%), ciprofloxacin (44.4%) and gentamicin (38.9%).⁴ As is evident from this, there is a high variation in the antibiotic susceptibility pattern against these pathogenic bacteria, and the reason for this might be linked to an antibiotics manufacture company, purchasing without a proper prescription, prescription without laboratory guidance, misuse and indiscriminate use, the study area and bacterial type, and resistance methods.

Conclusion

Ear infection affects all ages, but especially the young. The males had more infections than females. *Pseudomonas* spp showed a high positive growth percentage among the pathogenic gram-negative bacteria. Appear from antibiotic susceptibility pattern the azithromycin, gentamycin, piperacillin, ceftriaxone, cefepime, imipenem, ceftazidime and gentamycin showed activity against *Pseudomonas* spp, whereas trimethoprim, amikacin, imipenem and levofloxacin showed activity against *Serratia* spp, and levofloxacin and amikacin showed activity against *Klebsiella* spp.

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Conflict of Interest: None

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