

Journal homepage: wwwhttps://fezzanu.edu.ly/



# Prevalence of Bacterial Pathogens and Antimicrobial Resistance in Pediatric Otitis in Zintan, Libya.

\*Adel Jama<sup>1</sup> and Amani Al-shoglaf<sup>2</sup> and Fatima Al-Kasih<sup>2</sup>.

<sup>1</sup>*Departmen* of zoology, faculty of science, zintan university, Libya.

<sup>2</sup>Department of microbiology, faculty of science, zintan university, Libya

# A B S T R A C T

Bacterial infections causing otitis media are common in children and impact their ability to communicate and learn. These infections can lead to health and psychological complications, underscoring the need for medical intervention. The significance of this study lies in its investigation of bacterial infections in children with ear infections at healthcare centers in Zintan city, from May to August 2023. A total of 100 samples were collected from one week to five years, including 54 females and 46 males. Ear swabs were taken using sterile cotton swabs, followed by bacterial culture on various media. The identification of bacterial species was conducted through Gram staining and biochemical tests. Antibiotic susceptibility was assessed using the modified Kirby-Bauer disk diffusion method. The study identified 85 bacterial isolates, predominantly from the genera Staphylococcus, Klebsiella, and Escherichia. Gram-positive bacteria comprised 68.23%, with Staphylococcus aureus being the most common at 35.26%. Formula feeding was associated with a higher prevalence of bacterial infections (42.55%) compared to breastfeeding (38.85%). Additionally, natural delivery showed a greater prevalence of bacteria (60%) than cesarean delivery (40%). Female had a higher infection rate (56.47%) compared to males (43.52%). Antibiotic susceptibility testing revealed that Meropenem was highly effective (100% susceptibility), while Penicillin and Amoxicillin showed significant resistance (80%+). The findings of this study highlight the critical prevalence of antibiotic-resistant bacteria in pediatric ear infections, emphasizing the need for regular monitoring and tailored treatment strategies to combat resistance effectively. The study underlines the importance of antibiotic stewardship in clinical settings to enhance treatment outcomes.

**Keywords:** Otitis media, Bacterial infections, *Staphylococcus aureus*, Antibiotic Susceptibility, children.

انتشار مسببات الأمراض البكتيرية ومقاومة المضادات الحيوية في التهاب الأذن لدى الأطفال في الزنتان، ليبيا. \*عادل مسعود جامع<sup>1</sup>، و أماني عبدالحميد الشقلاف<sup>2</sup>، فاطمة الكاسح<sup>2</sup> أقسم علم الحيوان، كلية العلوم، جامعة الزنتان، ليبيا. <sup>2</sup>قسم الأحياء الدقيقة، كلية العلوم، جامعة الزنتان، ليبيا

#### الملخص

تعد العدوى البكتيرية المسببة لالتهاب الأذن الوسطى شائعة لدى الأطفال، ولها تأثير سلبى على قدرتهم على

\* :Corresponding author

E-mail <u>adel.jama@uoz.edu.ly</u> 20 December 2024 - Received in revised form15 januare 2025 Accepted 8 februare2025







التواصل والتعلم. يمكن أن تؤدي هذه العدوى إلى مضاعفات صحية ونفسية، مما يبرز الحاجة إلى التدخل الطبي المناسب

المواد والطرق: تهدف هذه الدراسة إلى التحقيق من العدوى البكتيرية لدى الأطفال المصابين بعدوي الأذن في المراكز الصحية في مدينة الزنتان، ليبيا. حيث أجربت هذه الدراسة من مايو إلى أغسطس 2023. جمعت 100 مسحة أذن من الأطفال الذين تتراوح أعمارهم ما بين أسبوع وخمس سنوات، شملت 54 أنثى و 46 ذكرًا. أخذت العينات باستخدام مسحات قطنية معقمة، ثم زراعت على الأوساط الزراعية المختلفة. شخصت الأنواع البكتيرية المعزولة باستخدام صبغة جرام والاختبارات الكيميائية الحيوية و تم تقييم حساسيتها للمضادات الحيونة.النتائج:أظهرت الدراسة وجود 85 عينة بكتيرية، حيث كانت أغلبيتها من أجناس المكورات العنقودية، والكليبسيلا، والإشريكية. شكلت البكتيريا الموجبة لصبغة جرام أعلى نسبة 68.23%، في حين سجلت المكورات العنقودية الذهبية النسبة الأكثر شيوعًا 35.26%. وكان لرضاعة الصناعية الثركبير بزيادة انتشار العدوى البكتيرية (42.55%) مقارنةً بالرضاعة الطبيعية (38.85%). بالإضافة إلى ذلك، أظهرت الولادة الطبيعية انتشارًا أكبر للبكتيريا (60%) مقارنةً بالولادة القيصرية (40%). كانت نسبة الإصابة أعلى لدى الإناث (56.47%) مقارنةً بالذكور (43.52%). أظهر اختبار حساسية المضادات الحيوية أن Meropenem كان فعالًا جدًا (100% حساسية)، بينما أظهرت Penicillin و Amoxicillin مقاومة كبيرة أكثر من (80%). الخلاصة: أظهرت النتائج الانتشار الواسع للبكتيريا المقاومة للمضادات الحيوية في التهابات الأذن لدى الأطفال، مما يبرز الحاجة إلى المراقبة المنتظمة ووضع واستراتيجيات علمية للعلاجات المخصصة لمكافحة المقاومة بفعالية. كما تؤكد الدراسة على أهمية إدارة المضادات الحيوبة في البيئات السربرية لتحسين نتائج علاج المرضى. الكلمات المفتاحية: التهاب الأذن الوسطى،العدوى البكتيرية، المكورات العنقودية الذهبية، الأطفال, حساسية المضادات الحيوبة.

# introduction

Hearing loss can have significant consequences [1]. Limitations in communication abilities may impact personal relationships, hinder educational progress, affect interactions with health and other services, and restrict employment and career opportunities. Additionally, difficulties with speech and language can lead to stigmatization and increase the risk of poor psychological health and overall well-being. The total global economic burden of hearing loss surpassed \$981 billion [2]. According to the 2019 Global Burden of Disease Study (GBD), it is estimated that over 1.57 billion people experience some degree of hearing loss (greater than 20 decibels; GBD 2019 Diseases and Injuries Collaborators, 2020). Otitis media is an inflammation of the mucous membrane in the middle ear [3]. It is a common disease among children [4], caused by the accumulation of fluid behind the tympanic membrane due to blockage of the Eustachian tube. Early detection and treatment of otitis media is crucial to prevent complications and ensure the child's well-being. In children, this tube is more horizontal, shorter, and wider than in adults, and it consists of more flexible cartilage. This structure allows pathogens from the nose and upper respiratory tract to reach the middle ear more easily [2]. The primary bacterial pathogens responsible for acute otitis media are Streptococcus pneumoniae and Haemophilus influenzae, which are also



Journal homepage: wwwhttps://fezzanu.edu.ly/



commonly linked to sinusitis and pneumonia [2]. Acute otitis media is one of the most prevalent infectious diseases among children, particularly following the common cold, especially in preschool-aged children. Studies confirm that otitis media is one of the most widespread illnesses and a significant public health challenge globally. It results in a substantial number of doctor visits, with more than 20 million visits annually to healthcare centers in the United States alone. In developing countries, factors such as poverty, lack of education, limited medical expertise, and insufficient healthcare resources have significantly contributed to the increase in the number of affected individuals, leading to numerous complications, particularly among children [3]. Otitis media is a prevalent health issue across all age groups. Staphylococcus aureus and Proteus, spp. are the primary pathogens found in ear discharge. Gentamicin and norfloxacin are recommended as the preferred antibiotics for empirical treatment of otitis media [5]. Otitis media can be acute, lasting less than six weeks, or chronic, persisting for more than six weeks. Recurrent otitis media refers to three episodes of the condition within six months or four episodes within one year. There are numerous risk factors for otitis media in children, though some of these factors may vary depending on the type of otitis media [6].

Methodology (Materials and Methods

Specimen collection procedures

The present study conducted in the period from May1st to August31st, 2023, was characterized by a meticulous specimen collection process. A total of 100 samples were collected from children aged 1 week to 5 years, including 54 females and 46 males, who visited the Healthcare Centers in Zintan city, located in the northwest of Libya. Some of these children were suffering from ear pain, while others were asymptomatic. The collection of ear swabs using sterile cotton swabs and the utilization of a convenient sampling technique ensured the thoroughness of our data collection.

# Culture and Identification of bacterial species.

Each specimen was divided into two parts, one portion used for a primary Gram stain, while the other portion was used for culture inoculations onto MacConkey Agar, Eosin methylene blue Peptone water, Mannitol Salt Agar and Blood Agar (Oxoid, UK). These cultures were then incubated aerobically at 37°C for 24 to 48 hours [7]. Gram-positive isolates were undergone to catalase and coagulase tests, while Gram-negative bacteria were subjected to oxidase, Sulfur Indole Motility (SIM), urease production, and citrate utilization tests.

# Antibiotic susceptibility testing

The antibiotic susceptibility pattern of bacterial isolates was determined using the modified Kirby-Bauer disk diffusion method [8]. A swab from the isolated bacterial colonies was transferred to sterilized glass tubes containing 5 ml of peptone water and incubated at 37°C for 24 hours. A colony suspension with a concentration equivalent to a 0.5 McFarland solution was prepared for each identified isolate and inoculated onto Mueller–Hinton Agar (Himedia, USA). Appropriate antibiotic discs were placed onto the medium and incubated at 37°C for 24 hours. The antibiotic disksused were Oxacillin (OX) 10  $\mu$ g, Gentamicin (GM) 10  $\mu$ g, Nalidixic Acid (NA) 30  $\mu$ g, Vancomycin (VA) 10  $\mu$ g, Doxycycline (DO) 10  $\mu$ g, Amoxicillin (AMC) 30  $\mu$ g, Tetracycline (TE) 10  $\mu$ g, Penicillin (P) 10  $\mu$ g, Chloramphenicol (CHL) 30  $\mu$ g, and Meropenem (MEM) 10  $\mu$ g.



Journal homepage: wwwhttps://fezzanu.edu.ly/



The inhibition zones were measured and recorded in millimeters. The Multiple Antibiotic Resistance (MAR) index, a measure of the extent of antibiotic resistance in the isolated bacteria, was determined using the formula "A/B," where "A" represents the number of antibiotics to which the isolated bacteria exhibited resistance, and "B" denotes the total number of antibiotics assessed in this study [9].

# Statistical analysis

The data were rigorously evaluated using SPSS (Version 23) software, with the level of significance set at p<0.05. A chi-square test was employed to determine the prevalence of bacterial species and the relationship between bacterial species infection to gender and the type of feeding. Additionally, the chi-square test was used to compare the prevalence of bacterial species according to the type of delivery of the child. Statistical analysis played a crucial role in drawing robust conclusions from our study. **Results and discussion** 

In this study, the results revealed that 85 out of 100 specimens were found to contain various bacterial species classified into three major genera: Staphylococcus, Klebsiella, and Escherichia. These species were isolated using traditional bacterial culture methods and identified through biochemical tests following culturing on various media. The results of this study, as shown in Table (1) indicate that Gram-positive organisms constituted 68.23% of all bacterial isolates, while Gram-negative bacilli accounted for 31.77%. The results demonstrated the presence of multiple types of bacteria in the ear, which are responsible for causing ear infections. Three bacterial genera were isolated, with the dominant genus being Staphylococcus, accounting for 58 isolates at a prevalence rate of 68.23%. This high prevalence of Staphylococcus, with 30 isolates identified as Staphylococcus aureus at a rate of 35.26%, followed by 28 isolates of Staphylococcus epidermidis at 32.94%, underscores the urgent need to address this issue. The genus Klebsiella spp. were next, with 26 isolates at a prevalence rate of 30.58%. The least prevalent was the genus E. coli, with only one isolate at a rate of 1.18%. In the current study, the isolation and identification of bacteria obtained from patients' ear cultures were carried out by examining the colony morphology of the bacterial isolates, performing microscopic Gram stain analysis, and conducting biochemical tests [10]. This finding is in accordance with the study by Chow et al. [11], which reported that Gram-positive organisms constituted 56% of all bacterial isolates, while Gram-negative organisms made up 44%. The predominant organisms isolated were Staphylococcus aureus, Staphylococcus epidermidis, Klebsiella spp., and Escherichia coli. Staphylococcus spp. showed the highest prevalence at 65%, which is consistent with the findings of a previous study by [12]. The most frequently isolated species were *Staphylococcus aureus* (35.26%), *Staphylococcus epidermidis* (32.94%), Klebsiella spp. (30.58%), and Escherichia coli (1.17%). This finding aligns with studies conducted by [13]. in Nigeria, as well as Roland and Stroman [14], all of which reported that Staphylococcus spp. were the most frequently isolated bacteria in cases of otitis externa. Additionally, Brook [15] reported that the most common aerobic bacterium in the bacterial flora isolated from the external auditory canal was Staphylococcus epidermidis.







Table 1.Shows the isolated bacterial genera.

Bacterial genera	Number of isolates	Percentage (%)
Gram positive	58	68.23
Staphylococcus aureus	30	35.29
Staphylococcus epidermidis	28	32.94
Gram negative	27	31.77
Klebsiella spp.	26	30.59
E. coli	1	1.18
Total	85	100%

\*Chi-square analysis demonstrated a highly statistically significant difference in the distribution of bacterial genera between Gram-positive and Gram-negative groups ( $\chi^2 = 85.0$ , p < 0.0001, critical value = 7.815 at df = 3).p value< 0.05.

The results presented in Table (2) summarize the prevalence of isolated bacteria in relation to gender among children with ear infections. A total of 37 bacterial isolates were identified in males, accounting for 43.52% of the total samples, while 48 isolates were identified in females, representing 56.47%. Among the bacterial species, Staphylococcus aureus was the most prevalent in females (22.35%) compared to males (12.9%). Klebsiella spp. was more common in males (16.47%) than in females (14.11%). Notably, Escherichia coli was identified only in females, with a prevalence of 1.18%. These findings indicate a significant variation in the prevalence of isolated bacteria in relation to gender, with a notably higher rate in females compared to males. This finding is consistent with the results of Hassan and Adeyemi [16], who reported a higher prevalence in females (52.7%) than in males (47.3%). Furthermore, similar trends were observed in the study by Bashir et al. [17], who also reported a greater prevalence in females.

Table 2.	Prevalence of isolated bacteria in relation to gender.
----------	--

	Male		female		
	No. of identified	Percentage	No. of identified	Percentage	
Identified bacteria	bacteria.	(%)	bacteria	(%)	
S. aureus	11	12.90	19	22.35	
S. epidermidis	12	14.11	16	18.82	
K. pneumoniae.	14	16.47	12	14.11	
E. coli	0	0	1	1.18	
Total	37	43.52	48	56.47	



Journal homepage: wwwhttps://fezzanu.edu.ly/



\*no statistically significant difference between males and females in the distribution of identified bacteria.p value< 0.05.

Moreover, Hassan and Adeyem [16], have observed a higher incidence of ear infections in females (52.7%) compared to males (47.3%). The most frequently isolated bacteria in females were *Staphylococcus spp.*, with *Staphylococcus aurous* (22.35%) and *Staphylococcus epidermidis* (18.82%) being the most prevalent. In contrast, the most prevalent bacterium in males was *Klebsiella spp.* (16.47%). These gender-based differences in bacterial prevalence underscore the importance of developing genderspecific treatment strategies, as they suggest that the current one-size-fits-all approach may not be optimal for all patients.

The results in Table (3) indicate that the main bacterial species associated with otitis media in children are *Staphylococcus aureus*, *Klebsiella spp.*, *Staphylococcus epidermidis*, and *Escherichia coli*, with prevalence rates of 40%, 30%, 25%, and 5%, respectively. Notably, *Staphylococcus aureus* was identified as the most common cause of external ear infections in children, with a prevalence rate of 40%. This was followed by *Klebsiella spp. at* 30%, while *Staphylococcus epidermidis* accounted for 25%. These findings are consistent with the study by Hassan and Adeyemi [16], which reported that ear infections caused by *Staphylococcus aureus* reached 38.5%, with a prevalence of *Klebsiella spp. at* 15.4%. Similarly, a study conducted in Nigeria by Ilechukwu et al. [18], indicated that infants and young children are at the greatest risk of developing otitis media, with peak prevalence occurring between six and thirty-six months of age. Anatomical factors, such as the shorter and more horizontally oriented Eustachian tubes in children under the age of five years, facilitate the reflux of commensal organisms from the nasopharynx into the sterile middle ear, leading to congestion of the tube.

	Affected children		Healthy ch	ildren
	No. of identified	Percentage	No. of identified	Percentage
Identified bacteria.	bacteria	(%)	bacteria	(%)
S. aureus	8	40	22	33.85
S. epidermidis	5	25	23	35.38
K. pneumoniae	6	30	20	30.77
E. coli	1	05	0	00.00
Total	20	100	65	100

 Table 3.
 Name, number, and percentage of isolated and identified bacteria from ear of Infected children and Healthy children.

\* Chi-square test revealed no significant difference in the prevalence of identified bacteria between affected and healthy children, ( $\chi^2 = 3.84$ , p-value of 0.278, critical value = 7.815 at df = 3).(p > 0.05)

Based on the results obtained in this study, as shown in Table (4), the type of delivery has a clear impact on the prevalence of bacteria. The highest bacterial prevalence was observed in natural deliveries compared to cesarean deliveries, with rates of 60% and 40%, respectively. The study also revealed that *Staphylococcus aureus* is the most



Journal homepage: wwwhttps://fezzanu.edu.ly/



common cause of external ear infections in children, with a prevalence rate of 40%. This was followed by *Klebsiella spp.* at 30% and *Staphylococcus epidermidis* at 25%. *E. coli* was recorded with a prevalence rate of 5%. These findings have significant implications for pediatric care. They are consistent with the study by Hassan and Adeyemi [16], which showed that ear infections caused by *Staphylococcus aureus reached* 38.5%, while the prevalence of *Klebsiella spp.* was 15.4%. The potential transmission of certain bacteria from the mother to the newborn through the microorganisms in the vagina, as suggested by this study, aligns with the findings of Mändar and Mikelsaar [19], which indicated a significant relationship between the level of bacterial contamination and the bacterial content in the vagina.

	Natural de	livery	Cesarean delivery		
	No. of identified	Percentage	No. of identified	Percentage	
Bacterium species	bacteria	(%)	bacteria	(%)	
S. aureus	10	11.76	20	23.52	
S. epidermidis	22	25.88	06	07.05	
K. pneumoniae	18	21.18	08	09.41	
E. coli	01	01.18	00	00.00	
Total	51	60.00	34	40.00	

Table 4. Bacterial Prevalence in Relation to Delivery Type

\*Chi-square analysis revealed a statistically significant difference in the distribution of bacterium species between natural and cesarean deliveries ( $\chi^2 = 14.50$ , p = 0.0023, critical value = 7.815 at df = 3).p value< 0.05.

Based on the results of this study, as shown in Table (5), which illustrates the relationship between different types of feeding and the prevalence of various bacteria, it was found that formula feeding recorded the highest percentage (42.55%), followed by breastfeeding (38.85%), and finally mixed feeding (18.82%). The study indicated that the highest prevalence of bacteria was found in children who were fed formula milk, which was higher than in those who were breastfeed. This finding is consistent with previous studies that indicate exclusive breastfeeding until the age of four months, followed by partial breastfeeding, is associated with a significant reduction in rates of respiratory and gastrointestinal diseases in infants [20,21]. These implications underscore the importance of our research in understanding and promoting pediatric health.

Table 5.Bacterialprevalence in relation to the feeding type.

	Natural breast feeding		Artificial feeding		Mixed	feeding
Bacterium species	No. of identifie	Percentag e (%)	No. of identifie	Percentag e (%)	No. of identifie	Percentag e (%)





Journal homepage: wwwhttps://fezzanu.edu.ly/

	d		d		d	
	bacteria		bacteria		bacteria	
S. aureus	3	03.52	22	25.88	5	5.88
S. epidermidis	20	23.52	4	04.70	4	4.70
K. pneumoniae	10	11.76	10	11.76	6	7.05
E. coli	000	00.00	0	00.00	1	1.18
Total	33	38.82	36	42.35	16	18.82

\* Chi-square analysis indicated a statistically significant difference in the distribution of bacterium species across different feeding types ( $\chi^2 = 31.13$ , p < 0.0001, critical value = 12.592 at df = 6). p value< 0.05.

#### Antibiotic susceptibility testing

The results presented in Table (6) indicate that Meropenem (Mem) demonstrated the highest efficacy, with a 100% susceptibility rate across all tested bacterial isolates. This underscores its potential as a highly effective treatment option for infections caused by these bacterial strains. Gentamicin (Gn) and Chloramphenicol (Chl) also exhibited excellent activity, with susceptibility rates of 95% and 97%, respectively. These findings suggest that these antibiotics remain potent choices for the management of bacterial infections in the studied population. In contrast, Vancomycin (Va) and Doxycycline (Do) showed slightly lower, but still substantial, susceptibility rates of 87%. While effective, these antibiotics may not be as universally reliable as Meropenem. The data for Ciprofloxacin (Cip) revealed a moderate susceptibility rate of 54%, indicating that a significant proportion of bacterial isolates exhibit resistance. This suggests that Ciprofloxacin may not be the optimal first-line therapy in this context, and its use should be carefully considered based on specific susceptibility profiles. Antibiotics such as Oxacillin (Ox) and Tetracycline (Te) displayed higher resistance rates, with 44% and 40% of isolates being resistant, respectively. This level of resistance raises concerns about their effectiveness and urgently suggests the need for alternative therapeutic options or combination therapy when these agents are considered. Finally, Amoxycillin (Amc), Penicillin (P), and Nalidixic acid (Na) showed notably high resistance rates (80% or higher), rendering them largely ineffective against onto the tested bacterial isolates. These findings strongly discourage the use of these antibiotics as monotherapy for treating infections within this bacterial population. Shows the antibiotic susceptibility testing of the isolated bacteria. Table 6.

Disc code	Potency	Susceptible	Intermediate	Resistant
Mem	10	85 (100 %)	0	0
Chl	30	83 (97 %)	2	0
Gn	10	81 (95%)	0	4
Va	10	74 (87 %)	0	11
Do	10	74 (87%)	0	11
Cip	5	46 (54 %)	12	27
Ox	10	38 (44 %)	0	47
	Disc code Mem Chl Gn Va Do Cip Ox	Disc codePotencyMem10Chl30Gn10Va10Do10Cip5Ox10	Disc codePotencySusceptibleMem1085 (100 %)Chl3083 (97 %)Gn1081 (95%)Va1074 (87 %)Do1074 (87 %)Cip546 (54 %)Ox1038 (44 %)	Disc code         Potency         Susceptible         Intermediate           Mem         10         85 (100 %)         0           Chl         30         83 (97 %)         2           Gn         10         81 (95%)         0           Va         10         74 (87 %)         0           Do         10         74 (87%)         0           Cip         5         46 (54 %)         12           Ox         10         38 (44 %)         0





Journal homepage: wwwhttps://fezzanu.edu.ly/

Antibiotic	Disc code	Potency	Susceptible	Intermediate	Resistant
Tetracycline	Te	10	34 (40 %)	0	51
Penicillin	Р	10	22 (25 %)	0	63
Nalidixic acid	Na	30	22 (25 %)	0	63
Amoxicillin	Amc	10/20	18 (20 %)	2	66

The data presented in Table (7) highlights the Multiple Antibiotic Resistance (MAR) indices of various bacterial species isolated from ear infections. *S. aureus* demonstrated a high MAR index of 0.64, indicating substantial resistance to multiple antibiotics, including ciprofloxacin (CIP), penicillin (P), nalidixic acid (NA), ciprofloxacin (CN), oxacillin (OX), amoxicillin-clavulanic acid (AMC), and tetracycline (TE).

This significant resistance underscores the need for careful antibiotic stewardship, a responsibility that we all share in the fight against antibiotic resistance. S. epidermidis exhibits a lower MAR index of 0.36, with resistance to penicillin (P), nalidixic acid (NA), amoxicillin-clavulanic acid (AMC), and tetracycline (TE). Although this resistance is relatively moderate, it still suggests a need for monitoring and potentially adjusting treatment regimens based on susceptibility profiles. Klebsiella spp. the highest MAR index of 0.72, reflecting considerable resistance to ciprofloxacin (CIP), penicillin (P), vancomycin (VA), oxacillin (OX), doxycycline (DO), amoxicillinclavulanic acid (AMC), and tetracycline (TE). This high level of resistance highlights the critical challenge of treating *Klebsiella* infections and may necessitate the use of more potent or novel antimicrobial agents. These results are consistent with those obtained by [21]. Escherichia coli demonstrates a MAR index of 0.36, with resistance to penicillin (P), vancomycin (VA), oxacillin (OX), and doxycycline (DO). While this index is lower compared to the other species, it still indicates significant resistance that could impact treatment efficacy. The results of this study are consistent with those of Es-Said et al. [22], who found that all bacteria were resistant to penicillin. They also observed that Escherichia coli isolates from children with ear infections were 100% sensitive to gentamicin, while Escherichia coli and Klebsiella spp. exhibited resistance to specific antibiotics, including ofloxacin and amoxicillin. Antibiotic resistance acquisition among bacterial species is a common phenomenon, achieved through several mechanisms such as vertical and horizontal gene transfer. Notably, conjugative transfer of plasmids carrying resistance genes is a key mechanism for the spread of resistance among bacteria [23]. Bacteria can develop various mechanisms to resist antibiotics, such as altering drug targets, blocking drug entry, expelling drugs through efflux pumps, or inactivating the drugs. To effectively select antibiotics for treating multidrug-resistant bacteria, it is crucial to understand and predict patterns of resistance [24].

No.	Bacteria specis	Resistance of antibiotics	MAR Index
1	S. aureus	CIP,P,NA,CN,OX,AMC, TE	0.64
2	S. epidermidis	P,NA,AMC,TE	0.36
3	K. pneumoniae	CIP,P,VA,OX,DO,AMC,TE	0.72
4	E. coli	P,VA,OX,DO	0.36

Table 7.Multiple antibiotic resistance index in identified bacteria isolated from the ear.



Journal homepage: wwwhttps://fezzanu.edu.ly/



OX. Oxacillin (10  $\mu$ g), GM. Gentamicin (10  $\mu$ g), NA. Nalidixic Acid (30  $\mu$ g), VA. Vancomycin (10  $\mu$ g), DO. Doxycycline (10  $\mu$ g), AMC. Amoxicillin (30  $\mu$ g), TE. Tetracycline (10  $\mu$ g), P. Penicillin (10  $\mu$ g), CHL. Chloramphenicol (30  $\mu$ g), MEM. Meropenem (10  $\mu$ g).

# CONCLUSION:

This study, which analyzed 100 samples from children in Zintan city, identified 85 bacterial isolates, predominantly belonging to the genera *Staphylococcus spp.*, *Klebsiella spp.*, and *E. coli*. The findings revealed a higher prevalence of Grampositive bacteria (68.23%), with Staphylococcus aureus being the most common isolate. Additionally, a notable difference in bacterial prevalence was observed between genders, with females showing higher rates. Antibiotic susceptibility testing indicated that Meropenem was highly effective, while significant resistance was noted for common antibiotics such as Penicillin and Amoxicillin. These results underscore the critical need for ongoing surveillance of antibiotic resistance patterns and the importance of targeted treatment approaches to effectively manage bacterial infections. The high MAR indices observed, particularly in *Klebsiella.spp.* and *S. aureus*, highlight the significance of clinical decisions in addressing antibiotic resistance to prevent treatment failures and improve treatment outcomes.

### DECLARATION

Funding This study received no funding.

### Acknowledgments

The authors express their sincere gratitude to the members of the Healthcare Centers in Zintan city for their support during their work. Special thanks to Dr. Zainab Ali Abolgqsem, the language editor who reviewed this manuscript, and Dr. Mohammad Al- Hensheri, and Dragana Mitić-Ćulafić,University of Belgrade-Faculty of Biology, who provided revisions and assisted in writing the manuscript.

#### **Authors' contributions**

Adel Jama, Amani Al-shoglaf, contributed to the sampling, data collection and conducted the laboratory analysis and writing up the manuscript. All authors read and approved the final edition of the manuscript and revision of the manuscript.

#### References

- 1. Olusanya, B. O., Neumann, K. J., & Saunders, J. E. (2014). The global burden of disabling hearing impairment: a call to action. Bulletin of the World Health Organization, 92, 367-373.
- 2. McDaid, D., Park, A. L., & Chadha, S. (2021). Estimating the global costs of hearing loss. International journal of audiology, 60(3), 162-170.
- 3. Bayehe, A.& Biadeglegne, F. (2009). Antimicrobial resistance patterns of Staphylococcus aureus and Proteus spp. isolated from otitis media at Bahir Dar Regional Laboratory, North West Ethiopia. Ethiop Med J. 47 (4):271-276.
- Nwokoye, N. N., Egwari, L. O., Coker, A. O., Olubi, O. O., Ugoji, E. O., & Nwachukwu, S. C. U. (2012). Predisposing and bacteriological features of otitis media. Afr. J. Microbiol. Res. 6 (3): 520-525.
- 5. Berman, S. (1995). Otitis media in children. New England Journal of Medicine, 332(23): 1560-1565.





Journal homepage: wwwhttps://fezzanu.edu.ly/

- 6. Assiri, K., Hudise, J., & Obeid, A. (2024). Risk factors for chronic and recurrent Otitis Media in children: a review article. Indian Journal of Otolaryngology and Head & Neck Surgery, 76(1): 1464-1469.
- Tiedt, N. J., Butler, I. R. T., Hallbauer, U. M., Atkins, M. D., Elliott, E., Pieters, M., Joubert, G. & Seedat, R. Y. (2013). Paediatric chronic suppurative otitis media in the Free State Province: Clinical and audiological features. South Africanmedical journal, 103(7): 467-470.
- 8. Bauer, A. W., Kirby, W. M., Sherris, J. C., &Turck, M. (1966). Antibiotic susceptibility testing by a standardized single disk method. American journal of clinical pathology, 45(4): 493-496.
- 9. Krumperman, PH. (1983). Multiple antibiotic resistance indexing of Escherichia coli to identify high-risk sources of fecal contamination of foods. Applied Environmental Microbiology, 46(1): 165-180.
- 10. Forbes B. A., Sahm, D. F.&Weissfeld A. S. (2007). Bailey and Scott's Diagnostic microbiology. 12th ed., Mosby Elsevier Company, USA.
- 11. Chow, V. T., Ho, B., Hong, G. S., & Liu, T. C. (1986). Bacterial and mycotic otological infections in Singapore. Epidemiology & Infection, 97(2): 385-392.
- Kisembo, P., Mugwanya, F., Atumanya, P., Othin, M., Oworinawe, R., Kagimu, B., Kisakye, A. &Bagambe, F. (2018). Prevalence of ear infections in first year children of primary schools in a Western Ugandan community. African Journal of Biomedical Research, 21(2): 117-122.
- 13. Akinjogunla, O. J.& Enabulele, I. O. (2010). Virulence factors, plasmid profiling and curing analysis of multidrug resistant Staphylococcus aureus and coagulase negative Staphylococcus spp. isolated from patients with acute otitis media. Journal of American science, 6(11): 1022-1033.
- 14. Roland, P. S.& Stroman, D. W. (2002). Microbiology of acute otitis externa. The Laryngoscope, 112(7): 1166-1177.
- 15. Brook, I. (1981). Microbiological studies of the bacterial flora of the external auditory canal in children. Acta Oto-Laryngologica, 91(3-4): 285-287.
- Hassan, O., & Adeyemi, R. E. (2007). A study of bacterial isolates in cases of otitis media in patients attending oauthc, Ile-Ife. African Journal of Clinical and Experimental Microbiology, 8(3): 130-136.
- Bashir, A., Amer Sabih, H., Ayub Ahmed Khan, A., Aamer, E., Shahid, F. &Sabeen Khurshid, Z. (2008). Microflora and antibiograms of discharging ears in a cross-section of population in Quetta, Balochistan. Pakistan Journal of Otolaryngology-Head and Neck Surgery. 24 (3): 56-59.
- Ilechukwu, G. C., Ilechukwu, C. G. A., Ubesie, A. C., Ojinnaka, C. N., Emechebe, G. O., &Iloh, K. K. (2014). Otitis media in children. Open Journal of Pediatrics, 4, 47-53.
- 19. Mändar, R., and Mikelsaar, M. (1996). Transmission of mother's microflora to the newborn at birth. Neonatology, 69(1), 30-35.
- 20. Monasta, L., Ronfani, L., Marchetti, F., Montico, M., Vecchi Brumatti, L., Bavcar, A., Grasso, D., Barbiero, C. & Tamburlini, G. (2012). Burden of disease caused by otitis media: systematic review and global estimates. PloS one, 7(4): e36226.



Journal homepage: wwwhttps://fezzanu.edu.ly/



- Atta, H. I., &Umar, F. F. (2021). Prevalence of otitis media in children attending a Primary Health Care Center in Samaru, Zaria, Nigeria. Ife Journal of Science, 23(1): 123-130.
- 22. Es-Said, I., Mahdoufi, R., Yagoubi, M., &Zouhdi, M. (2014). Isolation and antibiotic susceptibility of bacteria from otitis media infections in children in Rabat Morocco. J Biol AgricultHealthc, 4(28): 153-159.
- 23. Tao, S., Chen, H., Li, N., Wang, T., & Liang, W. (2022). The spread of antibiotic resistance genes in vivo model. Can J Infect Dis Med Microbiol. 3348695.
- 24. Chiş, A. A., Rus, L. L., Morgovan, C., Arseniu, A. M., Frum, A., Vonica-Țincu, A. L., and Dobrea, C. M. (2022). Microbial resistance to antibiotics and effective antibiotherapy. Biomedicines, 10(5), 1121.