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Original Article

PROFILES OF ANTIMICROBIAL RESISTANCE AND VIRULENCE GENES IN *Pseudomonas aeruginosa* FROM BURN WOUND INFECTIONS IN ERBIL, KURDISTAN REGION, IRAQ

Glena Qasim Faris[1,*], Karim Jalal Karim [2], and Hayman K. Mustafa[1]

[1]Department of Biology, Faculty of Science and Health, University of Koya, Koya KOY45, Kurdistan Region, Iraq.
[2] Department of Medical Laboratory Science, Faculty of Science and Health, University of Koya, Koya KOY45, Kurdistan Region, Iraq.

*Corresponding author, E-mail: glena.qasim91@gmail.com (Tel: +964-7729793601)

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ABSTRACT

P. aeruginosa is a remarkable opportunistic bacterium that presents a significant issue in the therapeutic context, particularly within burn units. This is due to its capacity to produce catastrophic infections and apparent resistance to a variety of antimicrobial medications. The current study was designed to characterize clinical isolates of this bacterium from individuals suffering infections in their burn wounds in emergency hospitals in Erbil, Iraq. Isolates were recognized and analyzed via conventional microbiological approaches, automated VITEK® 2 system and molecular diagnostic techniques. Thirty isolates were collected and confirmed by amplification of the *16S rRNA* gene. Antimicrobial drug susceptibility testing revealed high rates of resistance to several drugs, particularly ceftazidime, meropenem, gentamicin and ciprofloxacin. Molecular analysis revealed the existence of a large number of resistance genes coupled with efflux pumps, including *OprM* (90%), *MexB* (50%), *MexR* (43.3%) and *MexA* (36.6%). In addition, *PelA* and *PslA* genes associated with biofilm formation were found in all isolates (100%) and 30% of isolates, respectively. The findings underscore the critical need for ongoing molecular research on *P. aeruginosa* and the imperative to stop the dissemination of multidrug resistant species. The achievement of targeted infection handling strategies is essential.

KEYWORDS: Multidrug resistance, Burn infections, Biofilm, Efflux pumps, *P. aeruginosa* Molecular diagnostics.

1. INTRODUCTION

Pseudomonas aeruginosa (*P. aeruginosa*) is a Gram-negative, aerobic, non-spore-forming rod with an exceptional ability to live and persist in many environmental conditions. *P. aeruginosa* is a prevalent opportunistic infection in both hospitals and communities. Burns and wound infections pose a significant problem as they delay the healing process, encourage scar formation, and may result in bacteremia, sepsis, or organ failure syndrome (Younus *et al.*,2021). It is recognized as a main participant to hospital-acquired infections and a number of serious diseases has been associated to it for instance wound pneumonia, infections and keratitis (Stapleton & Carnt, 2012). Two main factors play role for the successful pathogenicity of *P. aeruginosa*: the first prominent factor is that it owns numerous virulence determinants that contribute to the establishment and survival of infections under diverse circumstances; and the second one is by using certain mechanisms for example β-lactamase production, efflux pumps, and mutations. It has been shown that *P. aeruginosa* has capacity to stand up against varied range of antibiotics utilized. *P. aeruginosa* has rod shape, measuring (1.0-5µm) length and (0.5-1µm) diameter. It has the ability to grow at different scale of temperature from 20 °C to 45 °C, and it is well known for their ability to produce certain pigments: pyocyanin, responsible for a blue coloration; pyoverdine, which imparts a green-yellow or brown-yellow hue; and, in some strains, pyorubin and pyomelanin, which produce red and black pigments, respectively. The bacterium does not ferment lactose but can utilize ammonia and acetate as sources of nitrogen and carbon, respectively, and can survive on minimal nutrients. Notably, *P. aeruginosa* exhibits exceptional resilience to environmental stresses, enabling its presence in virtually

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all types of soil and aquatic habitats worldwide (Varela *et al.,* 2023).

P. aeruginosa ranks as the third most prevalent pathogen responsible for nosocomial diseases, following both *Escherichia coli* and *Staphylococcus aureus*. It is one of the main causes of infections acquired in hospitals, particularly affecting individuals with Cystic Fibrosis, burn wounds, immunodeficiencies, in addition to persons undergoing mechanical ventilation (Karami *et al*., 2020). Burn is described as severe injuries of skin or further biological matters, typically resulting from exposure to heat, electrical currents, friction, chemical agents, or radiation (Abd El-Halim, 2021).

The most notable challenge to global public health is increasing resistance to antibiotic. It is revealed that antibiotic-resistant infections could cause up to ten million losses every year thru 2050 if effective interventions are not made to curb the current upward trend (Wu *et al*., 2024). To survive, *P. aeruginosa* develops a range of advanced mechanisms for resisting the inhibitor or bactericidal actions of antibiotics. *P. aeruginosa* possesses numerous virulence factors that compromise host defenses and facilitate infection. These factors encompass hemolysin production, pyocyanin production, gelatinase activity, and biofilm formation, which contribute to tissue injury and facilitate bacterial evasion of antibiotic treatment (Younus *et al*., 2021). The virulence factors, including flagella, pili, and lipopolysaccharides (LPS) that facilitate host adherence and colonization, proteases and toxins that cause tissue destruction, secretion systems that deliver effectors and toxins into the host, as well as quorum sensing and biofilm formation, enhance bacterial communication and resistance to treatment (Polse *et al.*, 2024).

These groups of bacteria protect themselves through strategies that include blocking antibiotic access into the cell, and actively expelling the antibiotics via efflux pumps, using enzymes that degrade the antibiotics or chemically altering their compounds, and modifying or safeguarding cellular targets to reduce antibiotic effectiveness (Darby *et al*., 2023). It is recognized that *P. aeruginosa* strains exhibit resistance minimally to four of the next antibiotics including (ceftazidime, imipenem, gentamicin, and ciprofloxacin) that are frequently isolated since nosocomial disease, especially in intensive care units. The efflux pumps and porin proteins, often involved in *P. aeruginosa* multidrug resistance (MDR). Four efflux pump systems of the Resistance Nodulation Division including (MexCD–OprJ, MexAB–OprM, MexXY–OprM, MexEF–OprN) are studied in detail system (Burcin *et al.*, 2012). The outer membrane porin OprD is also well studied. Genes that make up efflux pumps are usually co-located within operons of three genes. The initial gene codes for a membrane-fusion protein linked to cytoplasmic membrane (*MexA*, *MexC*, *MexE*, in addition to *MexX*). Furthermore, second gene codes for transporter proteins (*MexB*, *MexD* and *MexF*), which helps shuttle substrates across the inner membrane. An exterior proteins in the membrane (*OprM*, *OprJ*, and *OprN*) is encoded by the third gene to form the tripartite efflux system (Burcin *et al*., 2012).

The current research aims to accurately characterize *P. aeruginosa*, isolated from individuals with burn wound infection known to emergency hospitals in Erbil, Iraq, by using conventional microbiological and molecular diagnostic methods. Besides, the goal of the work was to recognize and describe key antimicrobial resistance genes in the isolates, thus augmenting our knowledge on the epidemiology and resistance of Pseudomonas. This is because it is recognized as a main participant to hospital-acquired infections and several serious diseases have been associated to it such as wound pneumonia, infections and keratitis.

2. MATERIAL AND METHODS

Sample Collection:

From September to November 2024, thirty burn swab samples were collected from burned persons in Erbil Emergency Hospital. The patients were 20 females and 10 males. On average, 40% of the total patients' body surface area burned. The samples were collected from different body parts including chest, legs, hands, and other affected areas.

Isolation and Growth Conditions:

Samples were inoculated immediately on MacConkey as well as nutrient agar and incubated in aerobic condition at 37°C overnight. The samples were then streaked on cetrimide agar medium (the selective medium for pseudomonads) and incubated at 37 °C for 24-48 hours (Khalid *et al.*, 2023) for microbial development and identification (Turki, 2024). VITEK system was employed for primary identification for the colonies individually and confirmation purposes. After identification, the confirmed colonies were kept at -80 °C in glycerol stock (Ali *et al.,* 2023).

Identification of the Isolates using VITEK2:

The assumed *P. aeruginosa* isolates were sent to Razi laboratory for confirmation. Final identification of *P. aeruginosa* was performed using the VITEK® 2 compact technique (bioMérieux, Canada), an automatic scheme established on biochemical profiling and AST (Antimicrobial Susceptibility Test) (Card: AST-N419) used. The identification was conducted in agreement with the company's guidelines; results were interpreted by means of the system's integrated software.

Extraction of Genomic DNA:

Pure bacterial colonies grown in liquid Luria-Bertani (LB) media; genomic DNA extracted using the Cell S.V small kit with GeneAll® ExgeneTM (Songpa-Gu, Seoul, Korea). To guarantee growth to the logarithmic phase, cultures were incubated overnight at 37°C while being constantly shaken, which is optimal for harvesting bacterial cells. The harvested cells were immediately processed for genomic DNA extraction. Using electrophoresis on a 0.8% agarose gel, the isolated DNA's purity and integrity were evaluated. Furthermore, a Nano Drop spectrophotometer was used to measure both concentrations and purity of DNA samples (Bakr *et al.*, 2022).

Table -1: Oligonucleotide Sequences of Primers Used in the Present Work.

Gene Name	Gene Primer Name	Primer Sequence 5'-3'	PCR Products (bp)	References
MexA	*MexA F*	CTCGACCCGATCTACGTC	503	(Jameel *et al.,* 2023)
MexA	*MexA R*	GTCTTCACCTCGACACCC		
MexB	*MexB F*	TGTCGAAGTTTTTCATTGAG	280	
MexB	*MexB R*	AAGGTCAC GGTGATGGT		
OprM	*OprM F*	GATCCCCGACTACCAGCGCCCCG	247	
OprM	*OprM R*	- ATGCGGTACTGCGCCCGGAAGGC		
MexR	*MexR F*	GCGCCATGGCCCATATTCAG	673	(Al-Mashhadani *et al.,* 2024)
MexR	*MexR R*	GGCATTCGCCAGTAAGCGG		
16S rRNA	*16S rRNA F*	GGGGGATCTTCGGACCTCA	956	(Altaai *et al*., 2014)
16S rRNA	*16S rRNA R*	TCCTTAGAGTGCCCACCCG		
PelA	*PelA F*	CATACCTTCAGCCATCCGTTCTTC	786	(Attallah AL-Mashhadani, *et al.*, 2024)
PelA	*PelA R*	CGCATTCGCCGCACTCAG		
PslA	*PslA F*	TCCCTACCTCAGCAGCAAGC	656	
PslA	*PslA R*	TGTTGTAGCCGTAGCGTTTCTG		

Gene Amplification Using PCR Technique:

Isolates were confirmed by polymerase chain reaction technique (Alpha, PCR max, U.K) to amplify genes responsible for biofilm and antibiotic-resistant based on the identification of a partial *16S rRNA* gene, as shown in Table (1). 20 µL of the master mix (Ampliqon, Denmark), 2.0 µL of every primer, 3.0 µL genomic DNA, and 40 µL of PCR water made up the PCR mixture. The PCR program was as follows: the cycles were different according to the amplified gene (Table 2). 1.5% agarose-gel-electrophoresis was run to recognize the PCR products (Hamasalih & Abdulrahman, 2020). The researcher independently optimized and developed the PCR amplification conditions for all target genes during this study.

Table -2: The PCR Conditions for Amplified Genes Studied in the Current Study.

Genes	Denaturation (temp / Duration)	Denaturation, Annealing and Elongation (temp. / duration)	No. of cycles	Final elongation (temp. / duration)
16S rRNA	95°C/5 minutes	95°C/ 40 seconds 58°C/ 45 seconds 72°C /60 seconds	25	72°C/10 minutes
mexA	94°C/3 minutes	94°C/ 30 seconds 57°C/ 45 seconds 72°C/ 60 seconds	32	72°C/10 minutes
mexB	94°C/3 minutes	94°C/ 30 seconds 57°C/ 45 seconds 72°C/ 60 seconds	32	72°C/10 minutes
mexR	94°C/3 minutes	94°C/ 30 seconds 57°C/ 45 seconds 72°C/ 60 seconds	32	72°C/10 minutes
oprM	95°C/10 minutes	95°C/ 15 seconds 55°C/ 10 seconds 72°C/10 second	40	72°C/5 minutes
pelA	95°C/5 minutes	95°C/ 40 seconds 58°C/ 45 seconds 72°C/ 60 seconds	35	72°C/10 minutes
pslA	95°C/5 minutes	95°C/ 40 seconds 58°C/ 45 seconds 72°C/ 60 seconds	35	72°C/10 minutes

3. RESULTS

Morphological and Cultural Characteristics:

Between September and November 2024, 30 *P. aeruginosa* isolates were taken from burned persons who were brought to Erbil Emergency Hospital. After being cultivated on MacConkey, nutrient, and cetrimide agar plates, isolates are incubated overnight at 37°C. Every strain showed strong growth on these media.

On Nutrient agar, the colonies appeared smooth, translucent, and large, exhibiting a low convex morphology with diameters ranging from 2 to 4 mm. The colonies emitted a characteristic fruity odor. Additionally, most isolates produced diffusible pigments, notably bluish-green pyocyanin, which is typical of *P. aeruginosa* (Figure 1A).

When cultured on MacConkey agar, the colonies were colorless, reflecting the organism's inability to ferment lactose. The colonies were large, flattened, round, and mucoid in texture. They emitted a distinctive odor reminiscent of corn tortilla or fruity odor similar to grapes. Some strains also produced characteristic pigments, including bluish pyocyanin and green pyoverdine, which imparted coloration to the surrounding medium (Figure 1B).

Cetrimide agar medium is selective medium to identify *P. aeruginosa*. On this medium, the colonies were smooth and mucoid, with raised centers and flat edges. They exhibited a fruity odor and ranged in color from yellow to green (Figure 1C).

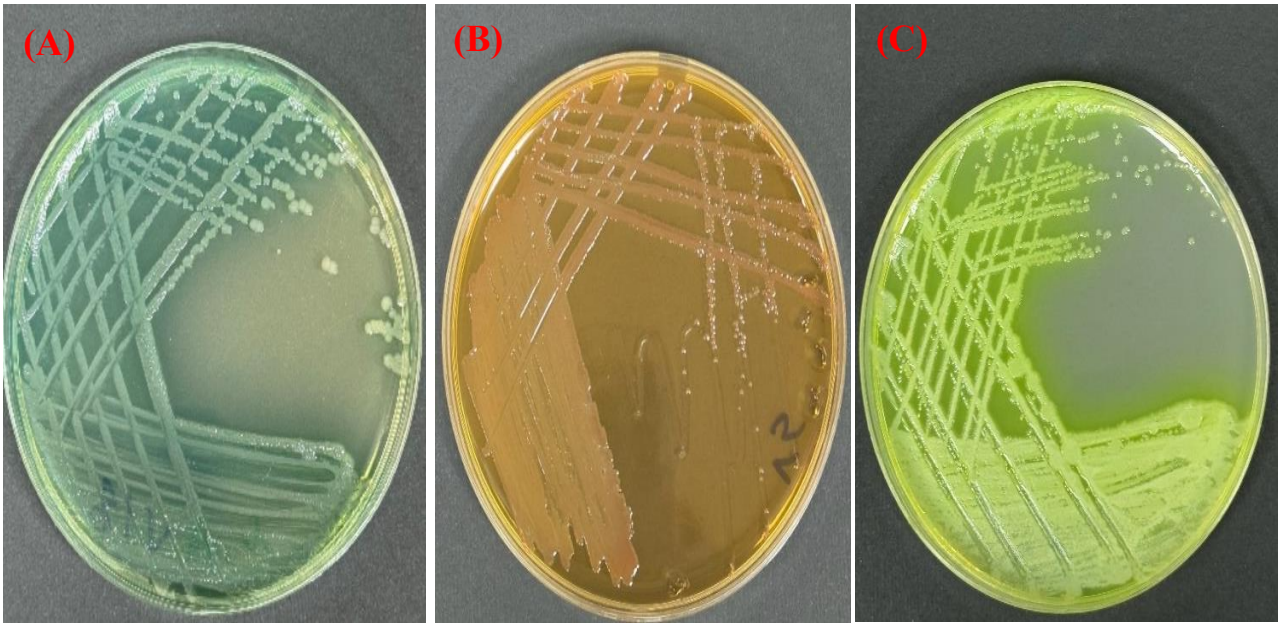

Figure-1: Cultural characteristics of the isolated bacteria (*P. aeruginosa)* from burned patients. The isolates were grown on Nutrient agar (A), MacConkey agar (B), and Cetrimide agar (C), respectively.

Identification of *P. aeruginosa* Using VITEK® 2 and Antimicrobial-Susceptibility Testing:

Identification of *P. aeruginosa* isolates was initially based on cultural characteristics and the presence of distinctive pigments. To confirm these preliminary identifications, every isolate was subjected to additional analysis utilizing the VITEK® 2 Compact system. A total of 30 isolates tested; all were identified as *P. aeruginosa* and the majority showed with 99% similarity. Subsequently, all isolates underwent antimicrobial susceptibility testing against a panel of eleven antibiotics to determine their resistance profiles. Ceftazidime, Ceftazidime/Avibactam, Piperacillin/Tazobactam, Ceftolozane/Tazobactam, Cefepime, Imipenem, Meropenem, Gentamicin, Amikacin, Colistin, and Ciprofloxacin were among the antibiotics that were evaluated. The detailed susceptibility results are presented in Table 3. It was shown that 56% of the strains showed resistance to ceftazidime. Piperacillin/Tazobactam resistance was present in about 45% of the samples; whereas, the remaining isolates were sensitive. Fewer than half of the isolates demonstrated resistance to the combination of Ceftazidime/Avibactam. Amikacin resistance was detected in only 40% of the strains. Notably, over 60% of the bacterial isolates were resistant to Ceftolozane/Tazobactam. In contrast, approximately 77% of the isolates remained sensitive to Cefepime, with the remainder showing resistance. Resistance rates to Imipenem and Meropenem were observed in less than half of the isolates. Among the antibiotics tested, Colistin demonstrated the highest effectiveness, with about 76% of the isolates being susceptible to it. All the isolated bacteria showed resistances to Gentamicin and Ciprofloxacin approximately at 60% and 66% respectively.

Table 3: The Results of Antibiotic Susceptibility Test of the Isolated *P. aeruginosa*.

Samples	Percentage	Piperacillin/ Tazobactam	Ceftazidime	Ceftazidime/ Avibactam	Ceftolozane/ Tazobactam	Cefepime	Imipenem	Meropenem	Amikacin	Gentamicin	Ciprofloxacin	Colistin
V1	94	S	S	S	R	S	S	S	I	R	R	I
V2	99	R	R	R	R	TRM	R	R	R	R	R	R
V3	98	R	R	R	R	I	R	R	R	R	R	R
V4	95	R	R	S	S	S	S	S	S	S	S	R
V5	97	R	R	S	S	I	S	S	S	S	S	I
V6	99	S	S	S	S	S	S	S	S	S	S	R
V7	99	S	TRM	S	R	S	S	S	I	S	R	R
V8	99	S	S	S	S	S	S	S	S	S	S	R
V9	99	S	S	S	S	S	S	S	S	R	S	R
V10	98	TRM	R	R	R	R	R	R	R	R	R	I
V11	99	R	R	R	R	R	R	R	R	R	R	R
V12	99	S	S	S	S	S	S	S	S	R	R	R
V13	99	R	R	R	R	R	R	R	R	R	R	R
V14	99	TRM	R	R	R	I	TRM	R	R	R	R	R
V15	99	S	S	S	S	S	S	S	S	S	S	I
V16	99	S	S	S	S	S	S	S	S	S	S	R
V17	99	S	S	S	R	S	S	S	I	S	R	R
V18	99	S	S	S	S	S	S	S	S	S	S	R
V19	95	S	S	S	R	S	S	S	I	S	R	R
V20	99	R	R	R	R	R	R	R	R	R	R	I
V21	99	R	R	R	R	R	R	R	R	R	R	R
V20	99	S	S	S	R	S	S	S	I	S	R	R
V21	99	R	R	R	R	R	R	R	R	R	R	R
V22	97	S	S	S	R	S	S	S	S	S	S	R
V23	95	R	R	R	R	R	R	R	R	R	R	I
V24	99	R	R	R	R	R	R	R	R	R	R	I
V25	93	R	R	S	R	I	R	R	R	R	R	R
V26	93	S	S	S	S	S	S	S	S	S	R	I
V27	97	R	R	S	S	I	S	S	S	S	S	I
V28	99	R	R	R	R	R	R	R	R	R	R	R
V29	93	TRM	S	TRM	S	S	S	S	S	R	R	R
V30	99	R	R	S	R	R	R	S	S	R	R	R
Total Resistance		14	17	12	18	10	13	13	12	18	20	23
Resistance percentage		46%	56%	40%	60%	33%	43%	43%	40%	60%	66%	76%

R= Resistant. **S**= Sensitive. **I**= Intermediate. **TRM**= Total Running Memory

PCR Amplifications of Genes:

The length of amplified *16S rRNA* gene fragments was 956 bp of universal primers. All the isolates showed a band of (956 bp) for *16S rRNA* gene after PCR amplification (Figure 2-A). Additionally, a set of specific genes for biofilm and antibiotic-resistance genes (*MexA*, *MexR*, *MexB*, *OprM*, *PelA*, and *PslA*) amplified and were detected. The *MexA* gene was specifically targeted and amplified using designed primers through PCR. Among 30 *P. aeruginosa* isolates tested, approximately 36% isolates exhibited a positive amplification band corresponding to the *MexA* gene. The amplified product size was confirmed to be 503 base pairs, consistent with the expected fragment length for this gene. This selective presence of the *MexA* gene in approximately one-third of the isolates suggests variability in the distribution of this efflux pump component among the tested Pseudomonas strains. The PCR amplification was validated by electrophoretic analysis on agarose gel, ensuring the specificity and accuracy of the detected band size (Figure 2B).

The *MexR* gene was specifically targeted and amplified using primers designed for PCR. Of the isolated *P. aeruginosa,* tested, only 43% isolates exhibited a positive amplification band corresponding to the *MexR*

gene. This finding indicates that the presence of the *MexR* gene varies among the *P. aeruginosa* isolates analyzed in the current work the resulting bands are shown in (Figure 2C). PCR was employed to amplify the *MexB* gene within genomic DNA using specific primers. The isolates tested, about 50% exhibited a visible band, indicating successful amplification of the target region. This suggests a limited presence or expression of the *MexB* gene within the tested bacterial cells the resulted band (Figure 3A).

The *OprM* gene was successfully amplified in all bulk isolates. Out of 30 *P. aeruginosa* isolates, 90% were found to carry the *OprM* gene; the resulting bands are shown in (Figure 3B). The *PelA* gene was detected successfully, and it was present in all the isolated strains, as shown in (Figure 3C). Out of 30 *P. aeruginosa* isolates *PslA* gene only 30% of was possessing the *pslA* gene (Figure 3D). Results of all amplified genes are presented in Table 4.

Table 4: The Table Provides Detailed Information About Molecular Detection of All Genes.

Gene's Name	Percentages of Positive Genes	No. of positive samples	Product Size
16S rRNA	100%	30	956bp
MexA	36%	11	503bp
MexB	50%	15	280bp
MexR	43%	13	673bp
OprM	90%	27	247bp
PelA	100%	30	786bp
PslA	30%	9	656bp

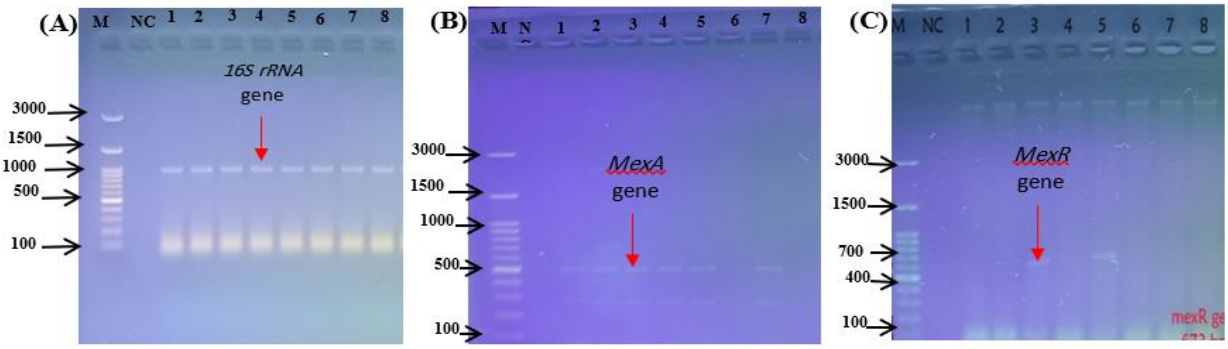

Figure 2: PCR amplification of *P. aeruginosa* target genes: *16S rRNA* gene (956 bp) (A), *MexA* gene (503 bp) (B), and *MexR* gene (673 bp) (C). Note; Lane 1-8: clinical isolates tested; Lane M: 100-3K bp DNA ladder; Lane NC: negative control (no template DNA).

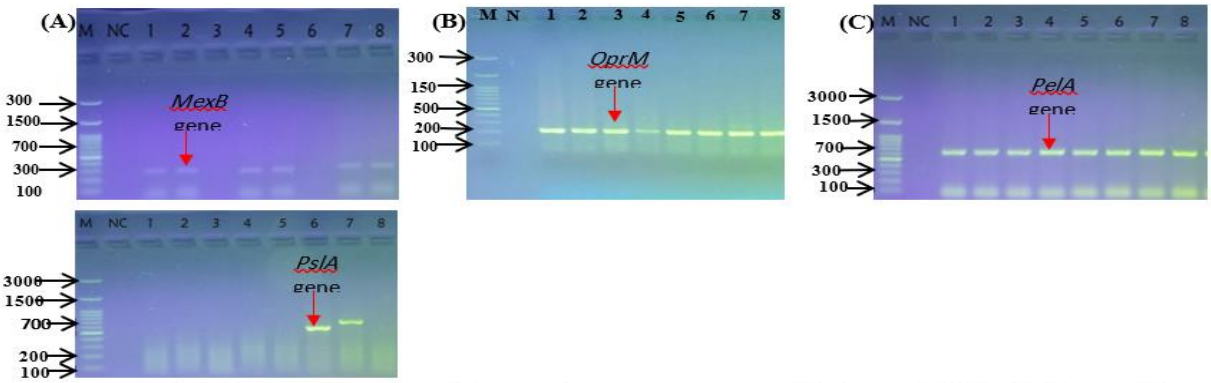

Figure 3: PCR amplification of *P. aeruginosa* target genes: *MexB* gene (280 bp) (A), *OprM* gene (247 bp) (B), *PelA* gene (786 bp) (C), and *PslA* gene (656 bp) (D). Note; Lane 1-8: clinical isolates tested; Lane M: 100-3K bp DNA ladder; Lane NC: negative control (no template DNA).

4. DISCUSSION

The morphological and genotypic traits of *P. aeruginosa* separated from infections of burn wounds at Erbil Emergency Hospital were examined in this study, focusing on antimicrobial resistance patterns, the existence of key virulence in addition to efflux-pump genes. The findings underscore the significant Multidrug-resistant bacteria represent a concern to public health. The identification of *P. aeruginosa* through both culture-based methods and the VITEK® 2 Compact system demonstrated high reliability. Out of 30 isolates, 25 were

confirmed with 99% confidence, consistent with the VITEK system's documented precision in differentiating closely related non-fermenting Gram-negative bacilli (Al-Bayati *et al.,* 2021; Hasan *et al*., 2020); Jaafar *et al.,* 2014) .

According to antimicrobial drug susceptibility tests, *P. aeruginosa* exhibited broad resistance against a diversity of antibiotics that were available, for instance β-lactams, aminoglycosides, fluoroquinolones. These results are consistent with earlier research showing that this bacteria acquired and possessed intrinsic resistance as a result of its efflux mechanism, active biofilm formation, and limited membrane permeability (Darby *et al*., 2023; Wu *et al*., 2024). Crucially, several isolates showed resistance to emerging medications e.g., ceftolozane, and ceftazidime, indicating the organism's rapid adaptation to antimicrobial pressure (Avakh *et al.,* 2023; Alsaadi, 2022).

Molecular characterization revealed that all isolates carried the *16S rRNA gene*, indicating that they were *P. aeruginosa*. The extensive discovery of resistance-related genes, particularly *OprM* (90%, 27/30 isolates), lends more credence to the idea that the RND family exocytosis outline shows major function in antibiotic resistance in these clinical bacteria. This aligns with earlier research that demonstrate the MexAB-OprM system is principally in charge of mediating defiance to series of antibiotics, particularly quinolones and β-lactams (Abbas *et al.,* 2018; Jameel *et al*., 2023; Wu *et al*., 2024).

Diverse exocytosis pump gene expression in clinical isolates is further demonstrated by the detection rates of the *MexA* (11/30), *MexB* (15/30), and *MexR* (13/30) genes. The lower detection rate of *MexA* compared to *MexB* and *MexR* may indicate that the expression of the exocytosis system is influenced by environmental or regulatory factors (Avakh *et al*., 2023). *MexR*, a repressor protein that controls expression of MexAB-OprM system, was present within less than half of isolated bacterial colonies. This might indicate mutational inactivation, which is usually associated with efflux pump overexpression and the resulting high level of drug resistance (Johnson & Church, 1999; Ozer & Savas, 2012).

Biofilm formation, one of *P. aeruginosa* primary virulence strategies, boosts the bacteria's viability and drug resistance. PelA's detection in all isolates is particularly noteworthy because it is required for the synthesis of Pel exopolysaccharides, which are crucial structural components of biofilm architecture (Colvin *et al*., 2012). This finding confirms earlier studies that this bacteria usually forms a high biofilm in burn injuries (Abdulla *et al.,* 2024; Fernández-Billón *et al.,* 2023; Ciofu *et al.,* 2019; Soares *et al.,* 2019). However, *PslA*, another gene involved in biofilm maturation, was only identified in nine of the thirty isolates, which may suggest that the isolates' methods for determining the composition of their biofilms differed.

P. aeruginosa exhibits a complex resistance mechanism that is revealed by the simultaneous identification of genes involved in biofilm and exocytosis. Interactions between these systems may lead to treatment failure and persistent infections. Furthermore, the presence of these genes in strains isolated in a small clinical setting suggests that the use of antibiotics by burn units may have accelerated the development and spread of resistant strains through local selective pressure (Sindeldecker & Stoodley, 2021).

The study's findings collectively bolster the clinical importance of tracking trends in antibiotic resistance through phenotypic susceptibility testing and molecular diagnostics. Resistance and virulence gene carriage are very common in *P. aeruginosa* isolated colonies from burned individuals in Erbil. This emphasizes how urgently strict procedures for infection mechanism, cautious antibiotic stewardship, investigation keen on alternative therapies like efflux pump inhibitors or anti-biofilm agents are needed.

CONCLUSION

Clinical *P. aeruginosa* strains that were sampled from burn injury patients in Erbil City, Iraq, have significantly multidrug resistance and genetic determinants for the development of a biofilm. The dissemination of genes that participate in the development of a biofilm and crucial efflux pump highly corresponds with resistance patterns that have been observed there. In the resistance against multidrug-resistant *P. aeruginosa* among the burn care units and high clinical risk units, the studies emphasize the serious need for the implementation of long-term molecular surveillance programs, the development of special infection control measures, and the adoption of novel methods for therapy.

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Ethical Statement:

The present study was evaluated and approved by the Research Ethical Committee of Koya University, Faculty of Science & Health, in accordance with established ethical guidelines (Approval Code: DBIO-5-2025).

Author Contributions:

G.Q. F., K.J.K., and H.K.M. have reviewed the final version to be published and agreed to be accountable for all aspects of the work.

Conflict of Interest:

The authors have no conflicts of interest to declare regarding this research.

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