Al-Mustansiriyah Journal of Science
ISSN: 1814-635X (print), ISSN:2521-3520 (online) Volume 33, Issue 3, 2022 DOI: http://doi.org/10.23851/mjs.v33i3.1129

Research Article **Open Access**

Correlation of Type 1 and Type 3 Fimbrial Genes with the Type of Specimen and the Antibiotic Resistance Profile of Clinically Isolated Klebsiella pneumoniae in Baghdad

Eman A. Muhsin[1*], Layla Abdulhemid Said[2], Sawsan Sajid Al-Jubori[2]

[1]Ministry of Science and Technology, Baghdad, IRAQ.
[2] Department of Biology, College of Science, Mustansiriyah University, Baghdad, IRAQ.

*Correspondent contact: eman2014bio@gmail.com

ArticleInfo

Received
12/01/2022

Accepted
12/03/2022

Published
25/09/2022

ABSTRACT

Klebsiella pneumoniae is a member of coliform bacteria that causes wide ranges of infections including circulatory, respiratory system, urinary tract infections (UTIs), and wounds infections. This study aimed to find the correlation between type 1 and 3 fimbrial genes expression with multidrug resistance (MDR) *K. pneumoniae* isolates towards antibiotics. Sixty clinical isolates of *K. pneumoniae* were collected from three main types of samples including blood, wound and burn swabs, and urine samples. The diagnosis was confirmed by VITEK-2 system and 16s *rRNA* housekeeping gene. The antibiotic sensitivity profile included 16 antimicrobial agents, with extended-spectrum beta-lactamase production. PCR technique was applied to detect four genes of type-1 fimbrial genes: (*usher-1*, *chaperon-1L*, *chaperon-1S*, and *fim-H1*), beside type-3 fimbrial genes: (*MrkA*, *MrkB*, *MrkC*, *MrkD*, and *MrkF*). The results showed that *K. pneumoniae* isolates were hundred percent (100%) resistant towards ampicillin, no resistance (0%) was recorded towards tigecycline and ertapenem, while the percentages of resistance for ceftazidem, cefepime, amikacine, and amipenem were 15%, 20%, 51.7%, and 50% respectively, and the isolates showed about (13-71%) resistance to the rest antimicrobials agents. The production of extended-spectrum beta-lactamase was in 40 (66.67%) of total the 60 isolates. There was no relationship according to the statistical analysis between the types of specimen with the antibiotic resistance rates. For fimbriae type 1 genes, the largest occurrence (90%) was reported in *Chaperon-1S* gene and the lowest one was in *Usher-1* gene (56.6%), while it was above 70% in *Chaperon-1L* gene and *fim-H1* gene of the total *K. pneumoniae* isolates. The percentages of type 3 genes *MrkA*, *MrkB*, *MrkC*, *MrkD*, and *MrkF* were: 28.3, 76.6, 85, 51.6, and 63.3% respectively. The type-1 fimbrial genes had no significant correlation among them; however, the type-3 fimbrial genes had significance in their presence at 0.01 and 0.05 levels, as they are located on the same Mrk operon. Finally, the correlation between type 1 and 3 fimbrial genes with the type of specimen and antibiotic resistance was not significant at all.

KEYWORDS: antibiotic resistance; *fimH1*; *usher-1* gene; *Mrk* genes; *Klebsiella pneumoniae.*

الخلاصة

تعد بكتريا الكليبسيلا الرئوية من البكتريا المعوية التي تسبب أنواع مختلفة من الإصابات منها تسمم الدم، إصابات الجهاز التنفسي، اضافة الى اصابات المسلك البولي والتهابات الجروح. تهدف هذه الدراسة الى ايجاد علاقة جينات الخمل نوع 1 و3 مع المقاومة المتعددة للمضادات الحيوية. تم اخذ عزل 60 عزلة من ال الكليبسيلا الرئوية في ثلاثة مجاميع من العينات وهي : الدم و،مسحات الجروح والحروق, وعينات الادرار والتي تم تأكيد تشخصيها باستخدام جهاز الفايتك-2 و استخدام الجين . *16s rRNA* تضمن فحص الحساسية للمضادات الحيوية 16 مضاداً حيوياً مع اختبار انتاج انزيم البيتالاكتاميز. تم تحديد وجود جينات الخمل نوع 1 (*Usher-1* و *chaperon-1L* و*chaperon-1S* و *fimH1*) ونوع 3(*MrkA* و *MrkB* و*MrkC* و *MrkD* و*MrkF*) بواسطة تقنية PCR. اظهرت النتائج مقاومة عزلات الكليبسيلا الرئوية بنسبة (100%) تجاه الامبيسيلين، مع عدم وجود مقاومة (0%) تجاه كل من التيجيسايكلين والارتابينيم. في المقابل كانت المقاومة تجاه السفتازديم، السيفسبيم، الاميكاسين والاميبينيم هي: (15% و20% و51.7% و50%) بالتعاقب وكان انتاج انزيم البيتالاكتاميز بنسبة 66.67% اي بواقع 40 من 60 عزلة بكتيرية. اثبتت نتائج التحليل الاحصائي عدم وجود علاقة معنوية بين وجود نوع العينة مع نسبة مقاومة المضادات الحيوية. فيما يخص نسبة وجود جينات الخمل نوع 1 فان أكبر نسبة سجلت هي (90%) للجين *chaperon-1S* بينما اقل نسبة هي (56.6%) للجين *Usher-1*، وكانت اكثر من (70%) لكل من *chaperon-1S* و *fimH1*. اما بالنسبة لجينات الخمل نوع 3 فكانت نسب وجود الجينات: (28.3% و 76.6% و 85% و 51.6% و63.3%) لكل من (*MrkA* و*MrkB* و*MrkC* و *MrkD* و*MrkF*) بالتعاقب. كانت التداخلات الاحصائية لوجود الجينات معنوية فقط ضمن جينات الخمل نوع 3 (عند مستوى

معنوية 0.01 و0.05) وذلك لوجودها على نفس الاوبرون *Mrk*، مع عدم وجود علاقة معنوية بين جينات الخمل نوع 1 و3 وبين المقاومة الميكروبية للمضادات الحيوية ضمن مجاميع العينات الثلاثة.

INTRODUCTION

Klebsiella pneumoniae is a multidrug resistant (MDR) bacterium that causes septicemia, lung infections, liver abscesses and urinary tract infection, which is considered as an opportunistic pathogen in both hospital and community-acquired diseases [1]. The adhesion properties of *K. pneumoniae* are generally mediated by type-1 and type-3 fimbria, which are consists of globular proteins that enables *K. pneumoniae* attach to the host cells as the first step in infectious process [2]. Type-1 fimbrial genes are essential for colonization, invasion and persistence of *K. pneumoniae* in the UTI. They are expressed from a chromosomal *Fim* gene cluster consisting of eight genes, while the type 3 belong to the chaperone-usher class of fimbriae and are encoded by five genes in addition to other genetic mechanisms in *K. pneumoniae (Mrk A, B, C, D* and *F)* [3].

Type 1 and type 3 fimbrial adhesins are mainly involved together with the antimicrobial MDR as there is a strong association that obviously explained the persistence of the MDR isolates for long time in the hospital environment and the difficulty of their eradication of *K. pneumoniae* [4].

Correlation of fimbriae type 1 and 3 with antibiotic resistance was well-characterized in *K. pneumoniae* [5]. Many pathogens initiate the colonization using fimbriae that mediate adhesion to host and environmental surfaces, facilitate invasion into the host tissues and promote bacterial interactions, which might suggest that the bacterium exhibits important resistance mechanisms that lead to the increasing pathogenicity and virulence [6]. *Klebsiella pneumoniae* harboring *fimH, mrkD* and other specific fimbrial genes that are sensitive to limited antibiotics, like meropenem, but not to others. These isolates of clinical *K. pneumoniae* are capable to develop MDR phenotype [7]. On the other hand, resistance genes encoded via some plasmids were evaluated with the presence of the fimbrial genes [8]. The current study aimed to detect the presence of type 1 and type 3 fimbrial genes in *K. pneumoniae isolates* and find their relationship with type of specimen and antibiotic resistance profile.

MATERIALS & METHODS

Sample collections

Sixty clinical isolates of *K. pneumoniae* were collected from three main types of samples including blood (20), wound and burn swabs (20) and urine samples (20) from three hospitals in Baghdad.

Bacterial Identification

The phenotypic examination for the isolates was first determined by culturing on selective media including MacConkey agar and blood agar media (Himedia, UK). VITEK-2 (Bio-Merieux, France) compact system for Gram-negative bacteria was used to detect the genus and species of the isolated bacteria via biochemical tests to ensure the primary identification of *K. pneumoniae*.

DNA Extraction

DNA extraction was performed (according to the instruction of DNA extraction wizard kit, Promega, USA), then concentration and purity of the extracted DNA were measured. The genotypic identification of the isolates was conducted using *16s rRNA* housekeeping gene (HKG) for all isolates. The sequences of *16S rRNA* primers were (5'AGAGTTTGATCCTGGCTCAG3') for the forward primer, and (5'TACGGTTACCTTGTT ACGACTT3') for the reverse primer [9]. PCR mixture consists of 12.5µl of green master mix (Promega/ USA), 3µl of template DNA, 1.5µl of each forward and reverse primers of *16S rRNA* gene and 6.5µl of nuclease-free water. After mixing by vortex, the mixtures placed in the thermocycler and all stages conditions were previously set in the thermocycler programme. Initial denaturation temperature was performed at 94 ℃ for 5 minutes for one cycle, followed by 40 cycles consisting of the 3 stages of PCR: Denaturation at 94 ℃ for 1 minute, annealing at 60 ℃ temperature for minute and extension at 72 ℃ for minutes. Finally, the final extension stage was done at 72 ℃ for 5 minutes and one cycle.

Antibiotic Sensitivity test by VITEK-2 Compact System

Antibiotic sensitivity against many antimicrobial agents was determined using VITEK-2 system by AST cards simultaneously with the detection of bacterial genus in this technique. In VITIK-2

system, the susceptibility towards different antibiotics had been interpreted according to CLSI (2021) for *Enterobacteriaceae* [10]. The antibiotics were used:- AMP: ampicillin (10µg), NIT: nitrofurantoin (300µg), CAZ: ceftazidim (30µg), CZ: cefazolin (30µg), CRO: ceftriaxone (30µg), CPM: cefepime (30µg), GM: gentamicin (10µg), P/T: pipracilin/tazobactam (100/10µg), FOX: cefoxitin (30µg), IMP: imipenem (10µg), SXT: sulfa/trimethoprim (1.25/23.75µg), AK: amikacin (30µg), CIP: ciprofloxacin (5µg), LVX: levofloxacin (5µg), TGC: tigecycline (15µg), ETP: ertapenem (10µg), in addition to the production of ESβLs: extended-spectrum beta lactamase (+/-) by tested *K. pneumoniae* isolates.

Molecular analysis of type 1 and 3 fimbrial genes

Fimbrial genes (type 1 and type 3) were amplified by PCR for 40 cycles with the same concentrations prepared for *16s rRNA* gene. All primers were designed in the current study by geneious software program, and their sequences are clarified in Table 1. Electrophoresis was then applied at 100 volt for 45 minutes using 1% agarose. The PCR product with forward primer or reverse primer for all detected genes were sent to Macrogen Company (South Korea) to be sequenced by genetic DNA analyzer using Sanger sequencing method. The results of sequence were analyzed using genius software depending on the comparison with the standard strain, which available in the National Centre for Biotechnology Information (NCBI).

Statistical analysis

Statistical analysis was performed using SPSS statistical package for Social Sciences (version 20.0 for windows, SPSS, Chicago, IL, USA). Qualitative data are represented as count and percentage. Chi-square test was used to test the relation of qualitative data. Spearman's rho correlation test was used to test the relation between qualitative data. *P-value* of <0.05 was considered statistically significant.

Table 1. Primers of fimbrial genes used in the study.

Gene symbol	Forward primer sequence (5′…3′)	Reverse primer sequence (5′…3′)	Size (bp)	Annealing temperature (°C)
Type 1 Fimbriae genes				
Usher-1	AATCGCCCCAGAGACTATATT	CTTGCTCTTTACTGGTTGACA	874	57.4
Chaperon-1L	AAACCATGCGCCAGAAAT	CGCATTTATTATTACGCCGC	361	52.5
Chaperon-1S	AAACCATGCGCCAGAAAT	AAATGTCGTACAGTTTGCGA	205	53.4
Fim-H$_1$	GCCAACGTCTACGTTAACCTG	ATATTTCACGGTGCCTGAAAA	180	53.4
Type 3 Fimbriae genes				
MrkA	GGCAGTTTTATTTTCTGACGG	GCACTAAACAGGATGACGTAA	358	57.4
MrkB	GGCCTTCATTTTTCTGATTGG	TTCATTTTTAGTTTTGCGGCC	346	53.4
MrkC	GAATTGTTGTAGCTGACCTGA	TCAACGCCTGAAAAACTATGT	1155	58.5
MrkD	GAACCCACATCGACATTCATA	CAGCAAACAACAAAGGATAGC	628	57.4
MrkF	TAAATATTCTGCGCTCCATCC	GGATTGCCGAAAAACACTATC	419	57.4

RESULTS AND DISCUSSION

Identification of *K. pneumoniae*

The colonies of *K. pneumoniae* characterized on blood agar media as circular, viscous, convex, smooth with distinct edges, lactose fermentation with no blood hemolysis. Bacterial diagnosis was confirmed using Gram-negative cards (ID-GN76) and confirmed using *16s rRNA* gene. Gel electrophoresis results showed *16s rRNA* band at 1500 bp. Figure 1: a and b illustrate the pairwise alignment of *16s rRNA* with standard strain of NCBI coded as (CP009775).

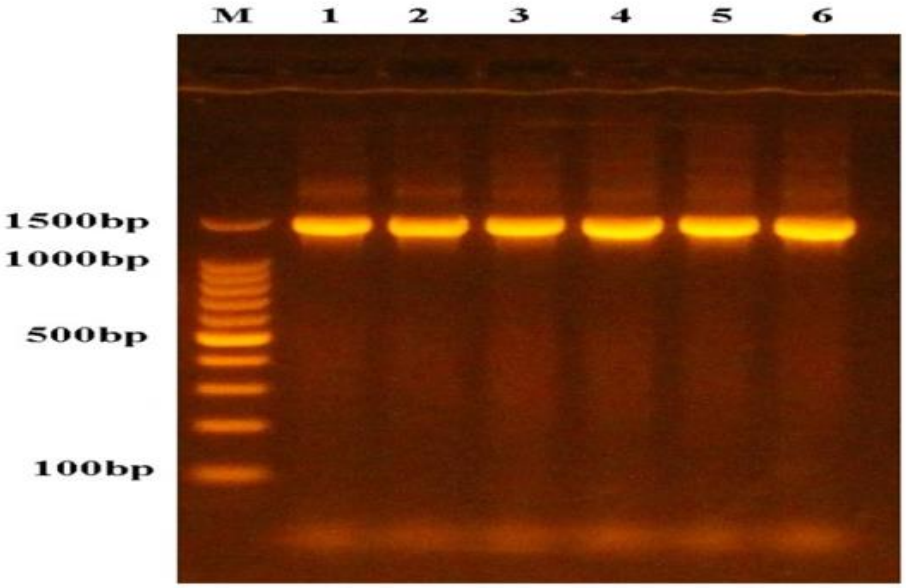
As clarified in the figures above, the pairwise identity with *K. pneumoniae* standard strain (CP009775) showed 99.8% identity. Bacterial PCR products had a positive result of *16s rRNA* gene confirming a *K. pneumoniae* homogenic data; by using DNA database from NCBI [11]. Moreover, molecular diagnosis using 16s *rRNA* gene was confirmed by other studies [12, 13].

Phenotypic detection of antibiotic resistance pattern by VITEK-2 system

As shown in Figure 2, *K. pneumoniae* isolates had a great resistance rate to the most antibiotics. The sensitivity percentage recorded towards ampicillin was the lowest (0%), while the highest sensitivity was towards ertapenem and tigecycline (100%).

On the other hand, *K. pneumoniae* isolates showed moderate sensitivity ranging between 40-71% for gentamycin, pipracillin/tazobactam, cefoxitin, imipenem, sulfa/trimethoprim, amikacin ciprofloxacin, and levofloxacin in a dramatic increasing pattern. The ESβL-negative isolates were (66.7%) of total isolates. For a less extent, sensitivity for ceftriaxone and cefepime was 20%, for nitrofurantoin was 15%, and finally it was 15% for both of ceftazidime and cefazolin. The results of this study are close to those obtained by [14, 15] regarding antimicrobial resistance rates.

(a)



(b)

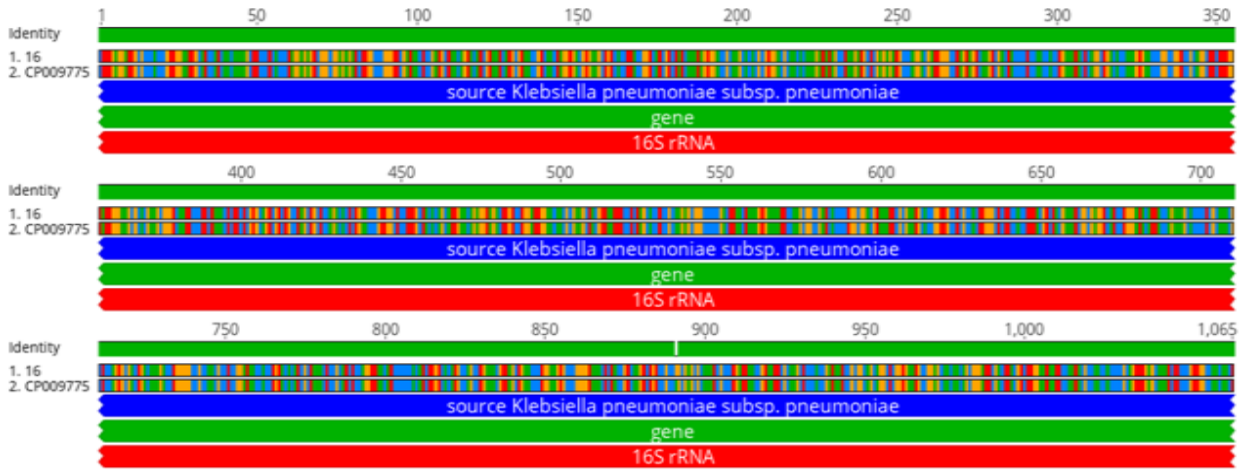


Figure 1. (a) Gel electrophoresis of PCR product with 1500 bp of *16s rRNA* gene in 1% agarose at 100V for 45 minutes. (b) Alignment of *16s rRNA* gene with the same gene of NCBI standard strains of *K. pneumoniae* (CP009775).

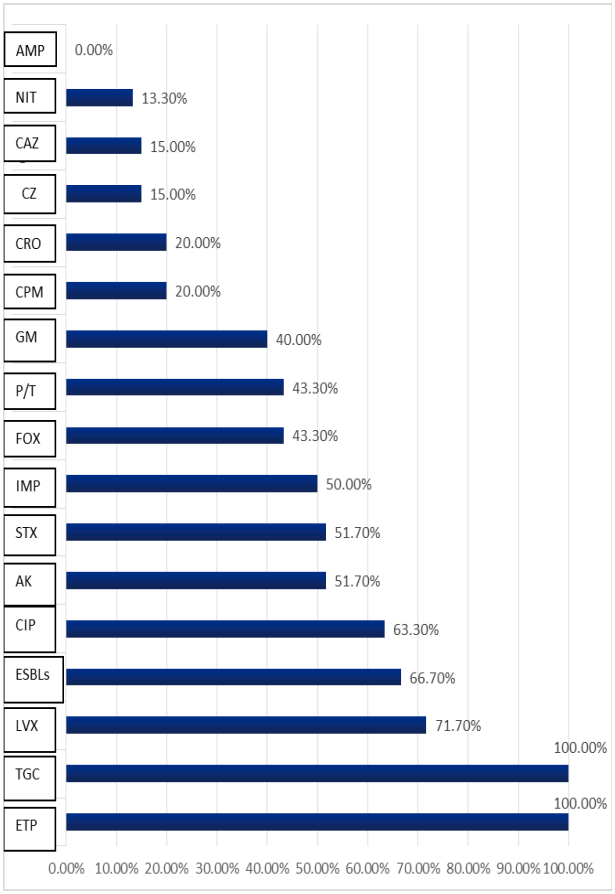


Figure 2. Antibiotic sensitivity of all *K. pneumoniae* strains. AMP: ampicillin (10µg), NIT: nitrofurantoin (300µg), CAZ: ceftazidim (30µg), CZ: cefazolin (30µg), CRO: ceftriaxone (30µg), CPM: cefepime (30µg), GM: gentamicin (10µg), P/T: pipracilin/ tazobactam (100/10µg), FOX: cefoxitin (3µg), IMP: imipenem (10µg), SXT: sulfa/trimethoprim (1.25/ 23.75µg), AK: amikacin (30µg), CIP: ciprofloxacin (5µg), ESBLs: extended-spectrum beta lactamase (+/-), LVX: levofloxacin (5µg), TGC: tigecycline (15µg), ETP: ertapenem (10µg).

For the antibacterial aspect, the ability of bacteria to develop resistance through diverse mechanisms or multiple pathways they possess by acquisition of new characteristics almost has been in random pattern which give the rise to elevating antibacterial resistance [16]; besides the development of invasive infections, like liver abscesses, endocarditis, meningitis, and septic arthritis, are considered as a serious issue which help in elevating MDR towards multiple antibiotics which struggle the antibacterial effect, but not towards newer drugs [17].

For the antibiotic resistance within the three specimen groups, there was no significant differences as shown in Figure 3 regarding to the source of bacterial isolation. The same result is recorded in the study of [13, 14] in which *K. pneumoniae* isolated from wounds, burns, sputum, urine, and ear swabs showed various resistance towards different antibiotics, which might be related to the disease progress and other underlying conditions [18, 19].

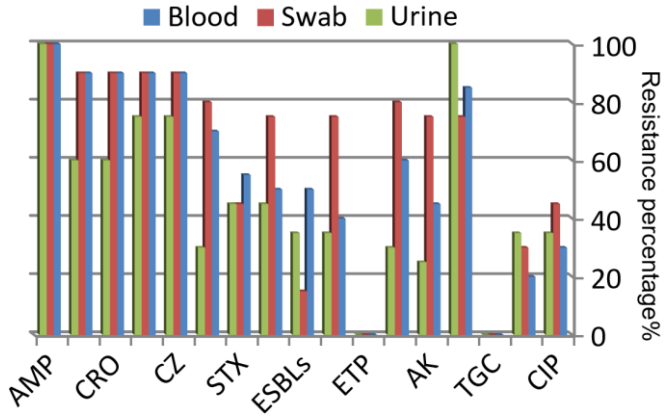


Figure 3. The antibiotic resistance within specimens' groups.

Detection of fimbrial genes

Gel electrophoresis results illustrated the precise size of PCR amplicons for fimbrial genes of type 1 and 3 as previously mentioned in Table 1. The electrophoresis was performed at 100V for 45 minutes in 1% agarose (Figure 4).

More than half (56.6%) of total *K. pneumoniae* isolates owned *Usher-1* genes, with a majority in urine samples. Many pathogens initiate a UTI using fimbriae that mediate adhesion to host and environmental surfaces, facilitate invasion into the host tissues and promote bacterial interactions to form biofilms by the swans of fimbriae assembled by chaperon-usher pathway.

As shown in Figure 5(a), the identity with the NCBI strain (CP071150) was 99.1%. The results are close to those [15, 20] of *usher-1* detection.

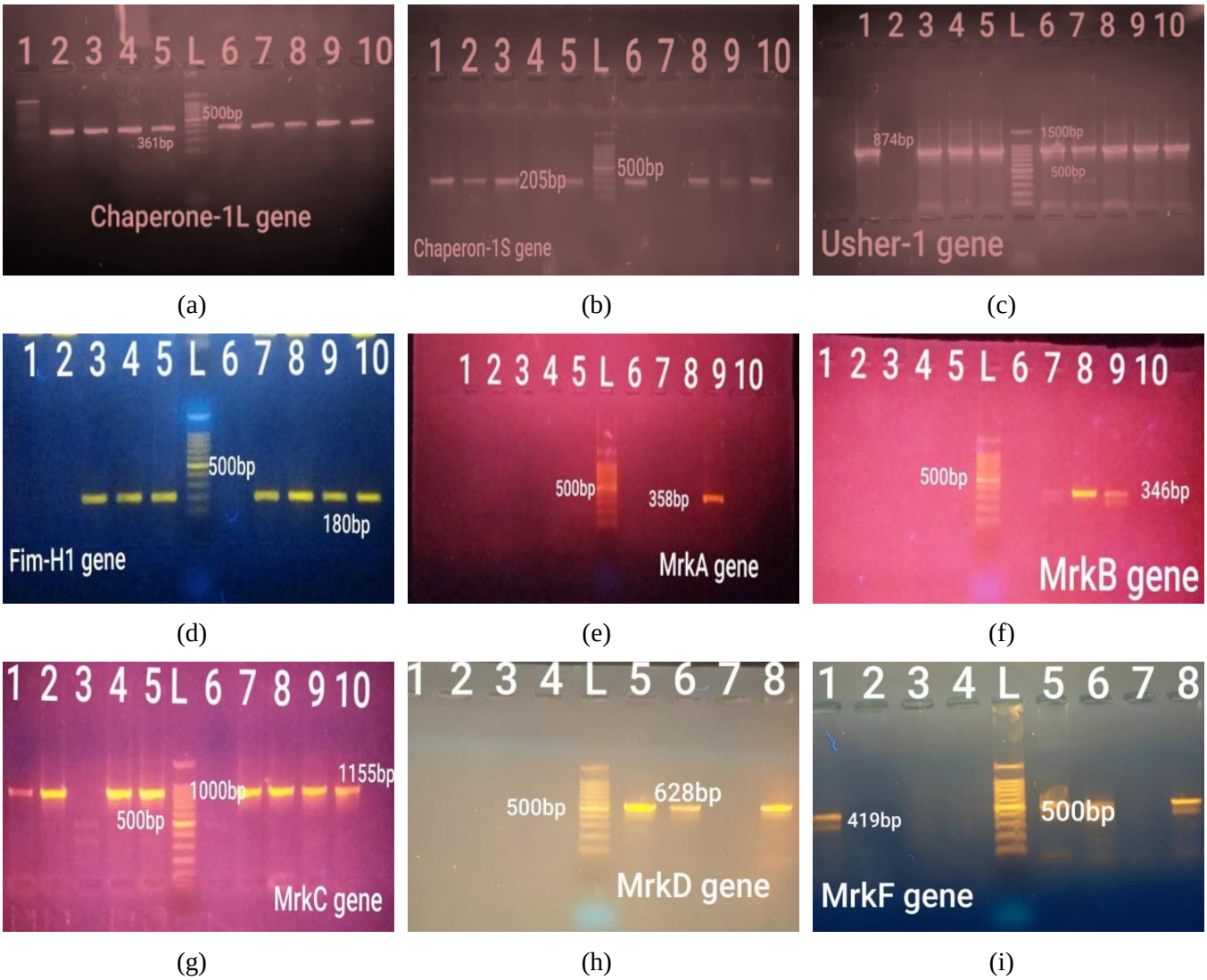


Figure 4. Gel electrophoresis in 1% agarose at 100V for 45 minutes of all studied genes. (a) *Chaperon-1L* gene (361bp). (b) *Chaperon-1S* gene (205bp). (c) *Usher-1* gene (874bp). (d) *FimH-1* gene (180bp) (e) *MrkA* gene (358bp). (f) *MrkB* gene (346bp). (g) *MrkC* gene (1155bp). (h) *MrkD* gene (628bp). (i) *MrkF* gene (419bp).

Chaperon- 1L gene, which is specific *to K. pneumoniae* adherence and pathogenicity, appeared in (71.6%) of bacterial isolates in this study. Figure 5(b) shows the identity of *chaperon-1L* gene with the standard NCBI strain (CP066855) which was 95.5%. The results seem close to that of [21, 22] about chaperon genes. *Chaperon-1S* gene, which is specific to *K. pneumoniae* adherence and pathogenicity, had the occurrence of 90% in bacterial isolates of this study. Figure 5(c) shows the *chaperon-1S* pairwise sequence alignment identity of 98.9% with NCBI strain (CP058940). The results are similar to those of [21, 22] about *Chaperon-1S* genes. The gene *fim-H$_1$* was detected in (78.3%) of all clinical isolates, most of them were isolated from blood samples (17 of 20 samples). *Fim-H$_1$* pairwise alignment with NCBI strain (CP068684) of 98.8% identity is clarified in Figure 5(d). The results are similar to those in the study of [12, 23]. All the studied of type 1 fimbrial genes were prevalent in urine samples and the highest percentage was for *Ch-1S* gene, followed by 80% for both of *Ch-1L* and *Fim-H$_1$* gene; and 65% for *Usher-1* gene. The high occurrence of type 1 fimbrial genes in urine samples explains how they together work to enable successful attachment, spread and persistence in the host urinary tract [3].

Al-Mustansiriyah Journal of Science
ISSN: 1814-635X (print), ISSN:2521-3520 (online) Volume 33, Issue 3, 2022 DOI: http://doi.org/10.23851/mjs.v33i3.1129

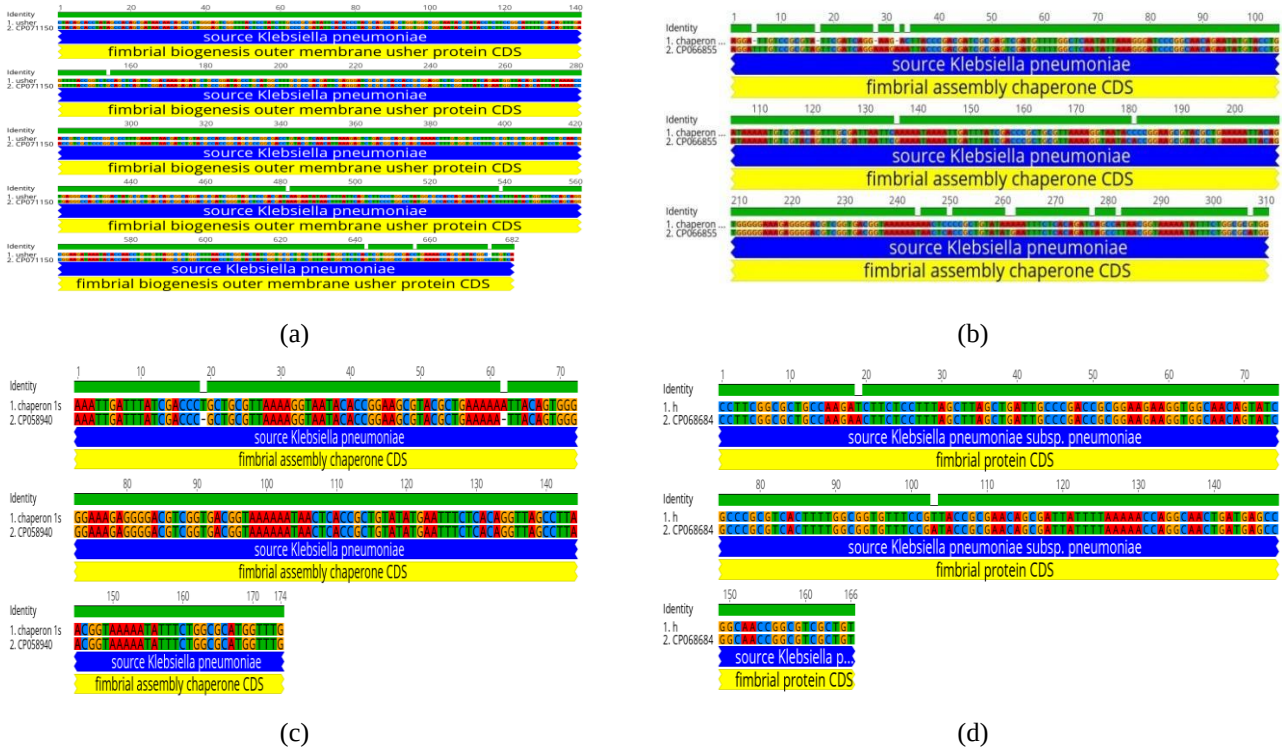


Figure 5. Gene pairwise sequence alignment of fimbrial type-1 genes with the same genes of NCBI standard strains of *K. pneumoniae*. (a) *Usher-1*alignment with (CP071150) standard strain. (b) *Chaperon-1L* alignment with (CP066855) standard strain. (c) *Chaperon-1S* alignment with (CP058940) standard strain. (d) *Fim-H$_1$* alignment with (CP068684) standard strain.

The percentages of type 3 fimbrial genes were: (28.3%, 76.6%, 85.0%, 51.6% and 63.3%) respectively. *MrkA* gene was present in 28.33% of total isolates. Figure 6(a) shows the identity is 98.4% with *K. pneumoniae* standard strain (CP043357). The results are close to the results of (3 and 19) about *MrkA* gene occurrence. *MrkB* gene was present in 76.66% of *K. pneumoniae* isolates. The pairwise sequence alignment was 97.4% identity with *K. pneumoniae* standard strain (CP073287) as shown in Figure 6(b). *MrkC* was present in 85% of *K. pneumoniae* isolates, with an identity of 94.4% in pairwise sequence alignment approach in comparison with NCBI strain CP047595 (Figure 6(c)). The results agreed with [23-25] who confirmed the occurrence of *MrkB* and *MrkC* genes. *MrkD* gene was present in 51.66% of *K. pneumoniae* isolates, with an identity of 99.8% with NCBI strain CP084497 (Figure 6(d)). The results are similar to the results obtained by [12, 8, 26]. *MrkF* gene was present in 63.33% of total isolates. Figure 6(e) below illustrated the pairwise sequence alignment identity of 99% with the NCBI strain (CP04335). The results matched with [23, 25] based on *MrkF* gene presence.

The prevalence of type 3 fimbrial genes showed the highest rates in urine *K. pneumoniae* isolates for *MrkC* (95%), *MrkF* (75%) and *MrkA* (40%), while *MrkB* gene was mostly prevalent in swabs (80%) and *MrkD* was (80%) in blood samples. Fimbria type 3 genes are synthesized by *K. pneumoniae* to facilitate the adhesion to cells including endothelial cells and uroepithelial cells. They also mediate adhesion to several cell types of the host tissues [6].

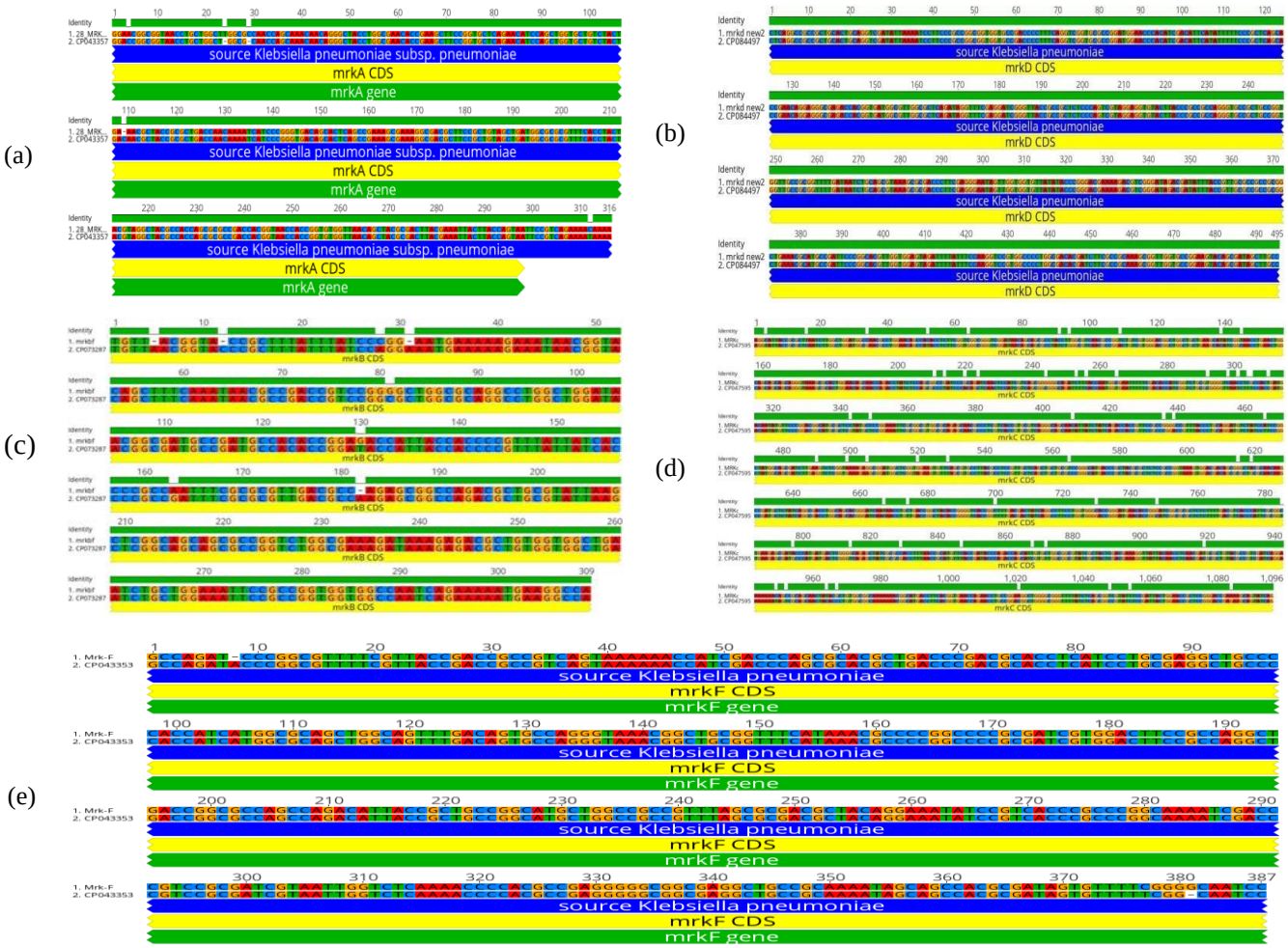


Figure 6. Alignment of fimbrial type-3 genes with the same genes of NCBI standard strains of *K. pneumoniae* (CP043357): (a) *MrkA*, (b) *MrkB*, (c) *MrkC*, (d) *MrkD*, (e) *MrkF*.

Relation of gene occurrence with the type of specimen

As clarified in Figure 7, there was no relation of genes with the specimen type and the presence of fimbrial genes. These agreed with [21, 22, 26, 27] who mentioned the absence of the relationship between gene occurrence and the specimen's source.

The absence of the association between the source of isolation and the genetic occurrence of *K. pneumoniae* isolates might be due to the high genetic diversity among the bacterial different clinical isolates in this study. The prevalence of one or more of the fimbrial genes among certain clinical *K. pneumoniae* isolates may contribute to the downregulation or overexpression of these genes [8]

Relation of antibiotic resistance with gene presence

As shown in Figure 8, the results are indicating the non-significant differences of antibiotic resistance rates among fimbrial gene. This result is similar to the results of [13, 28, 29]. The study of other entire factors and underlying diseases of the patients may suggest an expression to this negative relationship [19].

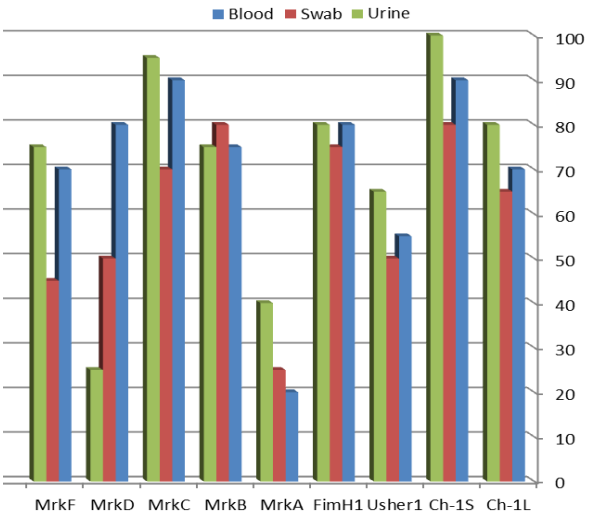


Figure 7. Gene occurrence percentages within specimen groups.

The characteristic of MDR *K. pneumoniae* is closely related to the antibiotic resistance genes encoded by plasmid and transferable genetic elements, and consequently *K. pneumoniae* continuous to accumulate these genes in the context of improper antibiotic use, resulting in the emergence of an extremely drug-resistant (XDR) strains; with the need to study further drug resistance mechanisms in *K. pneumoniae* clinical isolates [16].

Correlation between fimbrial genes 1 and 3 genes within all specimen groups

The correlations between each pair of genes were calculated by Pearson-correlation. Fimbrial genes occurrence of type 1 and 3 were correlated within the same type and with the genes of the other type of fimbriae. As shown in Table 2, the significance correlations were only among fimbrial type-3 genes, whereas there was no correlation among fimbrial tybe-1 genes or between them with the genes of type-3 genes. The findings of this study are close to those of [19, 30] who recorded there was no significance among type 1 fimbrial genes.

In this study, the positive covariant relationship (p-value is less than 0.01 or 0.05) among the type 3 fimbrial genes is corresponding to the fact that they arranged in the same transcriptional orientation as they are located on the same operon (*Mrk* operon), while lack of some type 1 fimbrial genes in *K. pneumoniae* leads to the overexpression of the other type-1 fimbrial genes with a negative inversive relationship [16].

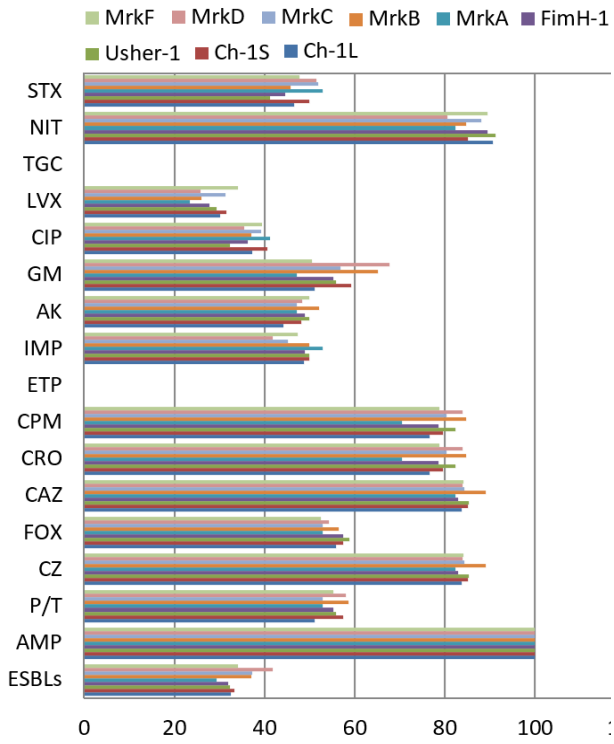


Figure 8. Antibiotics resistance rates within the studied genes.

Table 2. Correlations among fimbriae type 1 and 3 genes in *K. pneumoniae* isolates.

	MrkA	*MrkB*	*MrkC*	*MrkD*	*MrkF*	*Ch-1L*	*Ch-IS*	*Usher-1*	*FimH1*
MrkA	-	0.005**	0.011*	0.018*	0.018*	0.536	0061	0.336	0.239
MrkB	0.006**	-	0.016*	0.023*	0.023*	0.505	0.104	0.272	0.189
MrkC	0.011*	0.016*	-	0.050*	0.050*	0.413	0.057	0.270	0.306
MrkD	0.018*	0.023*	0.050*	-	0.0000**	0.697	0.104	0.426	0.169
MrkF	0.018*	0.023*	0.050*	0.0000**	-	0.697	0.104	0.426	0.169
Ch-1L	0.536	0.505	0.413	0.697	0.697	-	0.554	0.140	0.855
Ch-IS	0061	0.104	0.057	0.104	0.104	0.554	-	0.512	0.497
Usher-1	0.336	0.272	0.270	0.426	0.426	0.140	0.512	-	0.391
FimH1	0.239	0.189	0.306	0.169	0.169	0.855	0.497	0.391	-

*** Significant correlation at 0.05**
****Significant correlation at 0.01**

CONCLUSIONS

There was no significant relation of type 1, 3 fimbrial genes with the type of specimen or the antimicrobial resistance profile. This fact was dependent on the resistance profile of *K. pneumoniae* isolates in a limited concern; resulting

in the need to study the influence of other virulence factors in MDR.

ACKNOWLEDGMENT

Many thanks to the medical staff of hospitals in which samples were collected and to the college of Science, Mustansiriyah University, Baghdad, Iraq, for the support to complete this work (https://uomustansiriyah.edu.iq).

REFERENCES

[1] Oliveira, D., Forde, B. et. al., Antimicrobial Resistance in ESKAPE Pathogens. Clinical Microbiology Reviews. 2020: 33(3). https://doi.org/10.1128/CMR.00181-19

[2] Chatterjee, B., Puri, S. et al., Molecular Chaperones:Structure-Function Relationship and their Role in Protein Folding, 2018: Ch.8, Volume 13. https://doi.org/10.1007/978-3-319-74715-6_8

[3] Sonbol, F., El-Banna , T. et al., Conjugative plasmid mediating adhesive pili in virulent Klebsiella pneumoniae isolates. MedPub Journals. 2021: 3 (6), PP.4.

[4] Mahmoud, A., Biosysnthesis and characterization of some nanoparticles by using plants extracts and study their antimicrobial property against pathogenic bacteria isolated from wounds and burns. M.Sc thesis. College of Science. Diyala University, 2020.

[5] Scavuzzi, A., Maciel, M., et. al. Occurrence of qnrB1 and qnrB12 genes, mutation in gyrA and ramR, and expression of efflux pumps in isolates of Klebsiella pneumoniae carriers of bla KPC-2. Journal of Medical Microbiology. 2017, 66: pp. 477-484. https://doi.org/10.1099/jmm.0.000452

[6] Eghbalpoor, F., Habibi, M. et. al., Antibiotic resistance, virulence, and genetic diversity of Klebsiella pneumoniae in community and hospital acquired urinary tract infections in Iran. Acta Microbiologica et Immunologica Hungarica. 2019, 66(3): pp. 349-366. https://doi.org/10.1556/030.66.2019.006

[7] Caneiras, C. and Lito, L., Virulence and resistance determinants of Klebsiella pneumoniae isolated from a Portuguese tertiary university hospital centre over a 31-year period. Enferm Infecc Microbiol Clin. 2019, 37(6): pp. 387-393. https://doi.org/10.1016/j.eimc.2018.11.001

[8] Ragheb, S. Tawfick, M. El-Kholy, A. and Abdulall, A. Phenotypic and genotypic features of Klebsiella pneumoniae harboring carbapenemases in Egypt: OXA-48-like carbapenemases as an investigated model. Antibiotics J. 2020, 9:852. https://doi.org/10.3390/antibiotics9120852

[9] Hasan, S., Inhibition the expression of fimC, fimD, fimD genes in uropathogenic E. coli using TiO2 nanoparticles biosynthesized by probiotics bacteria. Ph.D Dissertation. College of Science. Mustansiriyah University, 2021.

[10] Clinical and Laboratory Standards Institut: A CLSI supplement for global application, 2021, 31sted. 40(1). USA.

[11] Shankar, C., Veeraraghavan, B. et. al., Whole genome analysis of hypervirulent Klebsiella pneumoniae isolates from community and hospital acquired bloodstream infection. BMC Microbiology. 2018, 18(6). https://doi.org/10.1186/s12866-017-1148-6

[12] Al-Musawi A., Genotypic and phenotypic typing of clinical Klebsiella pneumoniae local isolates. M.Sc. thesis. College of Science. Mustansiriyah University, 2018.

[13] Hamed, S. and Hasoon, N., Molecular characterization of carbapenemase-producing Gram-negative bacteria isolated from clinical specimens in Baghdad, Iraq. J. Pure. App. Microbiol. 2019, 13 (2): pp.1031-1040. https://doi.org/10.22207/JPAM.13.2.41

[14] Al-Oqaili, N., Molecular detection of β-lactamase production among klebsiella pneumoniae isolated from different clinical cases. Eurasia. J. Biosci. 2020, 14: pp. 35-41.

[15] Abbas, A., Hamed, S., Abdulsattar, B. and Mahmood, N., Molecular characterization of SHV-2a ESBL from clinical isolates of Klebsiella pneumoniae SLA55 in Baghdad, Iraq. Annals of R. S. C. B. 2021, 25(4): pp. 10993-11006.

[16] Wang, G., Zhao, G. et. al., The Characteristic of Virulence, Biofilm and Antibiotic Resistance of Klebsiella pneumoniae. Int. J. Environ. Res. Public Health 2020: 17. p: 6278. https://doi.org/10.3390/ijerph17176278

[17] Abbas, O., Mhawesh, A, and Al-Shaibani, A., Molecular Identification of Pathogenic Klebsiella Pneumoniae Strains Producing Biofilm. Medico-legal Update. 2020, 20: 3.

[18] Wasfi, R., Elkhatib, W. F. and Hossam M., Ashour Molecular typing and virulence analysis of multidrug resistant Klebsiella pneumoniae clinical isolates recovered from Egyptian Hospitals. Scientific Reports. 2016, 6:38929. https://doi.org/10.1038/srep38929

[19] Wyres, K., Lam, M. and Holt, K., Population genomics of Klebsiella pneumoniae Reviews. Nature. 2020:18. https://doi.org/10.1038/s41579-019-0315-1

[20] Ali, A. and Al-kakei, S., Virulence factors genotyping of Klebsiella pneumoniae clinical isolates from Baghdad. Reviews in Medical Microbiology. 2020, 30(1): pp. 36-46. https://doi.org/10.1097/MRM.0000000000000151

[21] Al-Kraety, I., Alquraishi, Z. and Alsadawi, A., Molecular Study of Fimh Gene in Klebisella Pneumoniae Isolated From Urinary Catheter Patients. Indian Journal of Forensic Medicine & Toxicology. 2020,14: 2.

[22] Fehan, A., Epidemiological study of bacterial urinary tract infection in Baquba City. M.Sc. thesis. College of Science. Diyala University, 2019.

[23] Al-Rubyaie, N., Detection of antiseptic resistance and ESBL genes in multidrug-resistant Pseudomonas aeruginosa and Klebsiella Pneumoniae from clinical and hospital environmentsources. M.Sc. Thesis. College of Science. Mustansiriyah University, 2020.

[24] Ali, A. and Al-kakei, S., Virulence factors genotyping of Klebsiella pneumoniae clinical isolates from Baghdad. Reviews in Medical Microbiology. 2020, 30(1): pp.36-46.
https://doi.org/10.1097/MRM.0000000000000151

[25] Luo, M., Yang, S. and Li, X. *et al.*, The KP1_4563 gene is regulated by the cAMP receptor protein and controls type 3 fimbrial function in Klebsiella pneumoniae. PLoS One. 2017, 12:7.
https://doi.org/10.1371/journal.pone.0180666

[26] Ong, C., Ulett, G. Mabbett, A. et. al., Identification of Type 3 Fimbriae in Uropathogenic Escherichia coli Reveals a Role in Biofilm Formation. J. Bacteriol. 2008,190(3): pp.1054-1063.
https://doi.org/10.1128/JB.01523-07

[27] Ali, Z., Determination of The Inhibitory Efficacy of Some Plant Extracts and Probiotics against Klebsiella pneumoniae Isolated from UTI Patients. Higher diploma thesis. College of Science. Tikrit University, 2019.

[28] Alwan, Z., Occurrence and Characterization of Genotoxin pks Gene in Clinically Isolates from Gram-negative bacteria. M.Sc thesis. College of Science. Diyala University, 2020.

[29] Aziz, R., Molecular detection of XDR and quorum sensing genes in local isolates of Acinetobacter baumannii and Klebsiella pneumoniae. Ph.D Dissertation. College of Science. Al-Mustansiriyah University, 2017.

[30] Al-Timimi, S., Persistence, and filaments formation in Klebsiella pneumoniae clinical Isolates. M.Sc. thesis. College of science. Mustansiriyah University, 2021.

How to Cite

E. A. Muhsin, l. A. Said, and s. S. . Al-jubori, “Correlation of Type 1 and Type 3 Fimbrial Genes with the Type of Specimen and the Antibiotic Resistance Profile of Clinically Isolated *Klebsiella Pneumoniae* in Baghdad”, *Al-Mustansiriyah Journal of Science*, vol. 33, no. 3, pp. 1–11, 2022.