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Original article

Distribution of *rmpA* gene and biofilm formation among hypervirulent *Klebsiella pneumoniae* isolates from Ain Shams University Hospitals

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ABSTRACT

Background: Hypervirulent *Klebsiella pneumoniae* (hv*Kp*) is an emerging pathogen causing life threatening infections. There are different determinants of virulence for these phenotypes like biofilm formation, large capsule, and siderophores. Many genes regulate these determinants as *rmpA* and *rmpA2*. **Aim of the work:** This study aims to detect the antimicrobial resistance profile, biofilm formation, and association between *rmpA* with hypermucoviscosity and biofilm formation among hypermucoviscous *K. pneumoniae* (hmv*Kp*) isolates. **Material and Methods:** Eighty *Klebsiella pneumoniae (K. pneumoniae)* isolates were collected from main microbiology laboratory of Ain Shams University Hospitals then subjected to phenotypic identification, antimicrobial susceptibility testing, biofilm formation ability, and detection of *rmpA* gene by PCR. **Results:** Eighty *K. pneumoniae* clinical isolates were tested, most of them were from sputum 30/80 (37.5%). 37/80 (46.25%) isolates were identified phenotypically as hmv*Kp* by string test while classic *K. pneumoniae* (c*Kp*) were 43/80 (53.75%). Biofilm formation was higher among hmv*kp* isolates than c*kp* A statistically significant difference of *rmpA* gene distribution among *K. pneumoniae* phenotypes were detected, the gene was detected in 31/37(83.8%) of hmv*Kp* isolates, and in 5/43(11.6%) of c*Kp* isolates respectively. Considering PCR as the gold standard, on comparing string test results with *rmpA* gene results, the sensitivity and the specificity of string were 86.11% and 86.36% respectively. **Conclusion:** Hmv*Kp* is being increased among *K. pneumoniae* clinical isolates with high rate of biofilm formation. The string test is a simple and rapid screening test of hmv*Kp* that compared with detection of *rmpA* gene as a confirmatory tool for hypermucoviscosity.

Introduction

Klebsiella pneumoniae (*Kp*) is one of the leading causes of healthcare acquired infections worldwide [1].

It is considered the second most common cause of bacteremia and a main pathogen causing hospital-acquired infection, particularly in patients with immunosuppression [2,3]. Higher mortality rate is associated with emergence of multidrug-resistant (MDR) *Kp* which reaches up to 40–50% [4].

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Hypervirulent *Klebsiella pneumoniae* (hv*Kp*), a new phenotype that emerged in last years and gained high clinical significance quickly, because it caused different types of infections with increased invasiveness among healthy immunocompetent hosts [5]. In comparison to classic *K. pneumoniae*, about half of infections caused by hv*Kp* occur in young healthy individuals with a high mortality rate that reaches 55%. Hyper-mucoviscosity (HMV) is used to differentiate between hvKp and c*Kp* which is determined by string test [6].

Virulence in hv*Kp* is mediated by different factors, but the capsule is considered the most important one. The genomic study highlights the presence of a large virulence plasmid that regulates hypermucoviscosity which includes the mucoid phenotype gene (*rmpA*) regulator. Additional virulence factors include biofilm formation. adhesins like (pili, fimbriae), urease production, and siderophores [7].

The *rmpA* (regulator of mucoid phenotype A) gene is mediated by plasmid. This gene is the regulator of the capsular polysaccharide synthesis, that confers a highly mucoviscous phenotype. In one study, it is clarified that removal of the *rmpA* gene can lead to reduction of virulence in mouse lethality tests by about 1000-fold as deletion of *rmpA* gene results in decrease of capsule synthesis which leads to decrease virulence of bacteria [8].

Formation of biofilm is also considered as one of very important virulence factors of hv*Kp* that add more to bacterial resistance to antibiotics. Different studies have demonstrated the relation between biofilm formation and virulence of hv*Kp*, biofilm was more pronounced among *rmpA* and *rmpA2* positive isolates than among isolates that lacking for these virulence factors [9].

Currently, the role of virulence factors, such as *rmpA* and hypermucoviscosity, in the biofilm of hmv*Kp* is poorly understood, so in this study we will search for the distribution of *rmpA* and biofilm formation among hmv*Kp*.

Aim of the work

The current study aims to determine the antimicrobial resistance profile, biofilm formation, and the association between *rmpA* with hypermucoviscosity and biofilm formation among hmv*Kp* isolates.

Material & methods

The present study was done on 80 *K. pneumoniae* isolates that were collected from different samples from Ain Shams University Hospitals, Main Microbiology laboratory, Cairo, Egypt during the period from January 2023 to June 2023. The study was approved by The Research Ethics Committee, Ain Shams University, Faculty of Medicine, and from MASRI (FMASU R03/2023).

Conventional identification, hypermucoviscosity assay and antimicrobial susceptibility testing

All isolates were identified by conventional microbiological methods by performing Gram stain, identifying lactose fermenting colonies on MacConkey agar and by biochemical reactions. The hv*Kp* is a variant phenotype of *Kp* and most of them show hypermucoviscosity. For identification of hypermucoviscosity, A string test is performed by testing the potential abilities of single colonies to stretch a string. if the formed string stretched more than 5 mm in length, the phenotype was considered hypermucoviscous [10] **(Figure 1).**

Then all isolates were tested for antimicrobial susceptibility by using disc diffusion method on plates of Muller-Hinton agar for the following antibiotics supplied from Oxoid, England: ampicillin (Am,10µg), amoxicillin/clavulanic acid (AMC,20/10 µg), ampicillin /sulbactam (SAM,10/10µg), piperacillin/tazobactam (TPZ,100/10 µg), cefeperazone/ sulbactam (CFS 75/10 ug) , ceftriaxone (CRO,30µg), cefoxitin (FOX, 10 ug), cefotaxime (CTX,30 µg), cefazoline (CZ,30µg), cefepime (FEP,30 µg), ceftazidime (CAZ,30µg), imipenem (IPM,10 µg), meropenem (MEM,10 µg), gentamicin (CN,10 µg), tobramycin (TOB, 10 ug), amikacin (AK,30 µg), ciprofloxacin (CIP,5 µg), levofloxacin (LEV,5 µg), ofloxacin (OFX, 5 ug) and trimethoprime-sulfamethoxazole (SXT, 1.25/23.75 ug). For urine specimens, Nitrofurantoin (F, 300 ug). *E. coli* ATCC 25922 was used as a quality control strain. The results were interpreted according to CLSI, 2023 [11].

Assessment of biofilm formation

A Microtiter plate was used to assess biofilm formation of all *Kp* isolates. An overnight culture in trypticase soy broth (TSB) was prepared and incubated at 37 °C for each isolate. Then, A total volume of 200 µL of cell suspension and TSB was inoculated in sterile microtiter plates (96 well plate) and a well for negative control was done by adding

200 ul of non -inoculated TSB. After that, at 37 °C incubation was done for 24 h.

After incubation 200 µL of phosphate-buffered saline (PBS) was used to wash wells gently for about three times then the wells were allowed to dry in an inverted position. The formed biofilm was stained by 0.1% crystal violet. After staining a 200 µL of distilled water was used to wash the plates for 3 times and left to dry in an inverted position. After all, the wells were dissolved in 200 µL of 5% isopropanol alcohol to solubilize the stained biofilm formed.

Measurement of optical density (OD) was done at 570 nm by using a microplate reader. Each well was tested for triplicate then, the mean OD was calculated. Optical density cut-off (ODc) was calculated as an average OD of negative controls + (3× standard deviation (SD) of negative controls).

Non-biofilm producer was reported if isolate in well showed OD ≤ ODc. while, the isolate was reported as biofilm producer either weak biofilm producer if 2 × ODc < OD ≤4 × ODc; moderate biofilm 2 × ODc < OD ≤4 × ODc; and strong biofilm producer if OD > 4 × ODc [12] **(Figure 2).**

Genotypic detection of *rmpA* gene

Extraction of bacterial DNA was done as by using extraction kit supplied from (QIAGEN, USA), for all isolates as per manufacturer's instructions then, *rmpA* gene amplification was done by using primers (*rmpA*-F 5'- ACTGGGCTACCTCTGCTTCA-3'; *rmpA*-R5'- CTTGCATGAGCCATCTTTCA- 3') supplied from Invitrogen. Amplification was done by using following parameters , an initial denaturation at (95°C for 5 min), followed by 35 cycles of (DNA denaturation at 95°C for 1 min), then primer annealing at (60°C for 1 min), then primer extension at (72°C for 5 min), then final extension (72°C for 7 min) according to [13]. Products of PCR were run on 2 % agarose gel, using ethidium bromide stain and seen under UV light and photographed and the *rmpA* gene was detected at 516 bp as shown in **figure (3).**

Statistical analysis

Data were statistically analyzed using statistical package for social science (SPSS) software version 25 was used. Qualitative data were expressed as frequency and percentages. The association between the qualitative data was assessed by chi-squared ($\chi 2$) test, and Fisher's exact test. *P* value < 0.05 was considered significant.

Figure 1. Hypermucoviscosity assay by string test. A and B showed positive string test results in *rmpA* positive hmv*Kp* isolates.

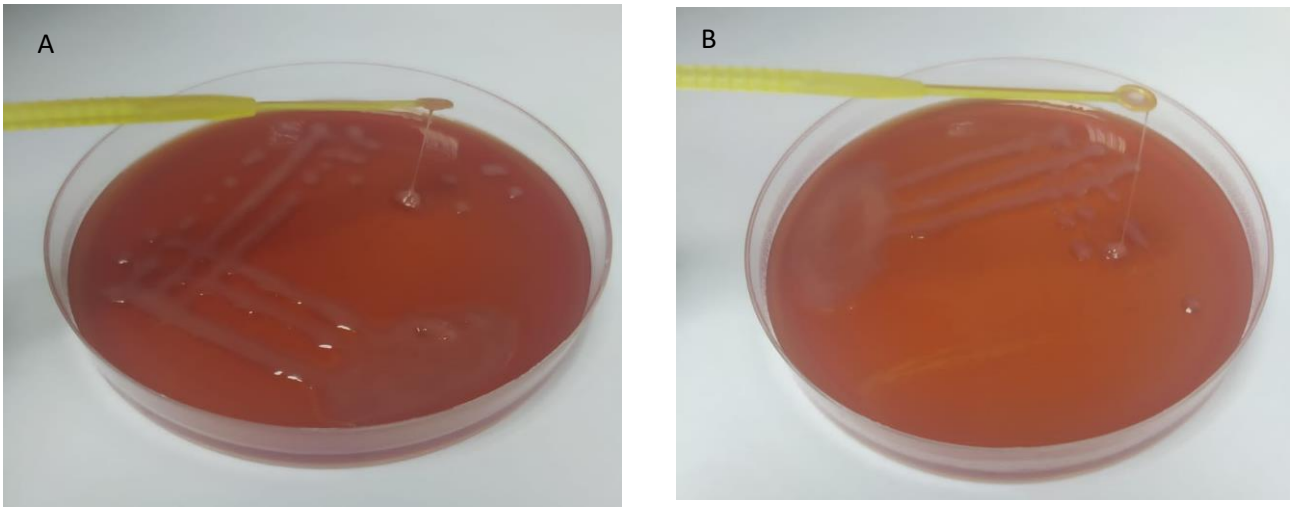


Figure 2. Detection of biofilm producing hmv*Kp* by using microtitre plate.
A- strong biofilm producer, B-weak biofilm producer and C- moderate biofilm producer.

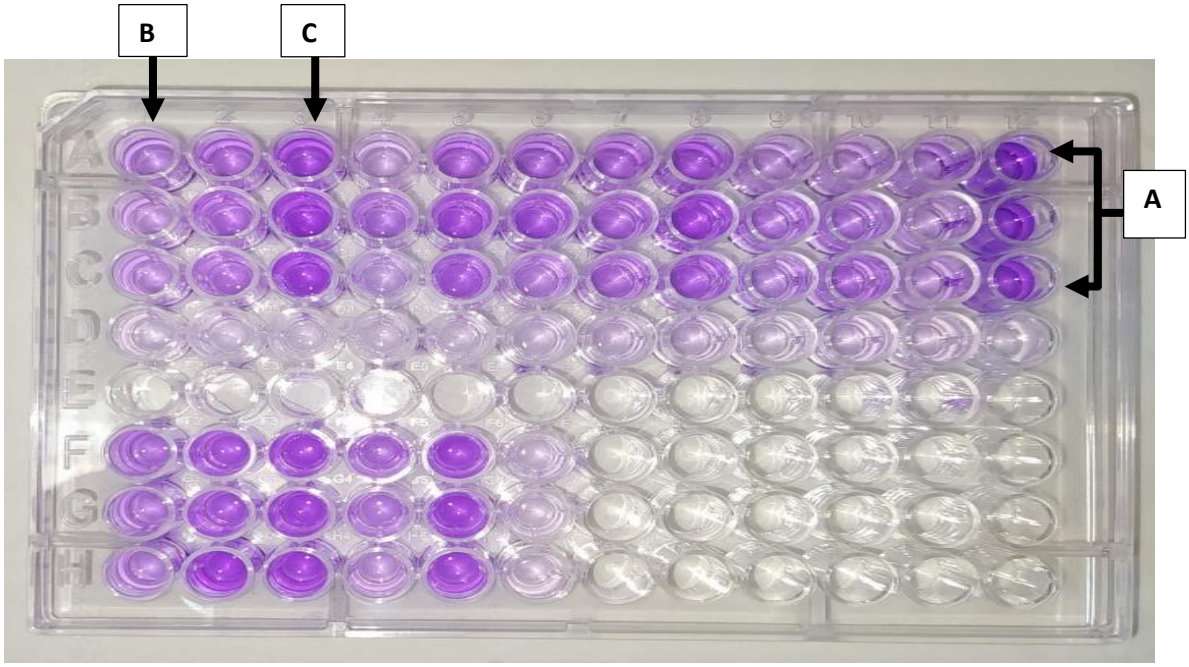
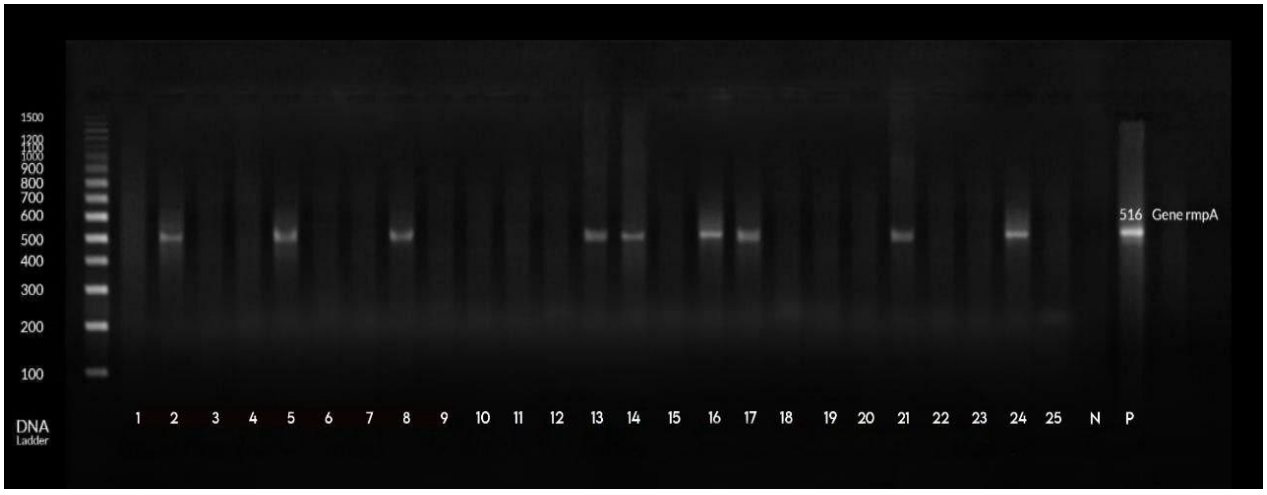


Figure 3. Positive detection of *rmpA* gene among *K. pneumoniae* isolates at 516 bp.



Results

The present study included 80 *K. pneumoniae* clinical isolates, 45/80 (56.5%) were retrieved from male patients while 35/80 (43.75%) were from females. Most of isolates were from sputum 30/80 (37.5%) followed by urine 26/80 (32.5%) then pus 24/80 (30%). Out of eighty, 37 (46.25%) isolates were identified phenotypically as hmv*Kp* by using string test and most of them were isolated from urine samples while c*Kp* were 43/80 (53.75%) and most of them were isolated from sputum samples as illustrated in **table (1).**

Regarding antimicrobial susceptibility of hmv*Kp* and c*Kp* isolates, all isolates (80) showed higher resistance to all beta-lactam antibiotics and cephalosporins but higher sensitivity towards imipenem and meropenem antibiotics with no statistically significant difference between hmv*Kp* and c*Kp* regarding the antimicrobial resistance pattern as described in **table (2).**

Biofilm formation was evaluated for hmv*kp* isolates, among the tested 37 isolates, there were 29/37 (78.4.%) isolates as biofilm producer and 8/37 (21.6%) isolates were non biofilm producers. Among the biofilm producers, the strong biofilm producers were 14/29 (48.2 %), the moderate biofilm producer were 12/29 (41.3%), and the weak biofilm producers were 3/29 (10.5%) but for c*Kp* isolates, among the tested 43 isolates, there were 23/43 (53.5%) isolates as biofilm producer and 20/43 (46.5%) isolates were non biofilm producers. Among the biofilm producers, 8/23 isolates (34.7%) were strong biofilm producers, 10/23 isolates (43.4%) were moderate biofilm producers, and 5/23 isolates (21.9%) were weak biofilm producers. There was statistically significant difference regarding biofilm formation in hmv*Kp* and c*kp* isolates **(Table 3, Figure 4)**

The distribution of *rmpA* gene was detected among all *Kp* isolates and revealed that among the hmv*Kp* clinical isolates, 31/37(83.8%) were harboring the gene while 6/37 (16.2%) were lacking the gene while among c*Kp* clinical isolates, 5/43(11.6%) were harboring the gene while it was absent in 38/43 (88.4%) of isolates. So, there was a highly statistically significant difference in the distribution of *rmpA* gene among hmv*Kp* and c*Kp* phenotypes **(Table 4).**

Also, the association between biofilm formation and *rmpA* gene among hmv*Kp* was evaluated and showed non statistically significant difference in the distribution of *rmpA* gene among hmv*Kp* isolates **(Table 5).** Considering PCR as the gold standard, on comparing string test results with *rmpA* gene results, the sensitivity was 86.11% and the specificity was 86.36% **(Table 6).**

Table 1. Distribution of hmv*Kp* and c*Kp* among different clinical samples.

Sample type	**Hmv*Kp* No. 37 (46.25 %)**		**c*Kp* No.43 (53.75 %)**	
Sputum	13	35.1%	17	39.5%
Urine	15	40.5%	11	32.3%
Pus	9	24.3%	15	40.5%

Table 2. Antibiotics susceptibility testing for Hmv*Kp* compared to c*Kp*.

Antibiotics	**Hypermucoviscus *K. pneumoniae* No. 37**			**Classical *K. pneumoniae* No.43**			**Sig**
	Resistant	**Intermediate**	**Sensitive**	**Resistant**	**Intermediate**	**Sensitive**	
	NO (%)	**NO (%)**	**NO (%)**	**NO (%)**	**NO (%)**	**NO (%)**	
Ampicillin	37 (100%)	0 (0%)	0 (0%)	43 (100%)	0 (0%)	0 (0%)	--
Ampicillin/ sulbactam	37 (100%)	0 (0%)	0 (0%)	43 (100%)	0 (0%)	0 (0%)	--
Amoxicillin/Clavulanic acid	37 (100%)	0 (0%)	0 (0%)	43 (100%)	0 (0%)	0 (0%)	--
Cefeperazone/ sulbactam	35 (94.6%)	0 (0%)	2 (5.4%)	38 (88.3%)	0 (0%)	5 (11.7%)	NS
Piperacillin/tazobactam	34 (91.9%)	0 (0%)	3 (8.1%)	37 (86%)	2 (4.6%)	4 (9.4%)	NS
Ceftazidime	35 (94.6%)	0 (0%)	2 (5.4%)	40 (93%)	0 (0%)	3 (7%)	NS
Ceftriaxone	33 (89.2%)	0 (0%)	4 (10.8%)	40 (93%)	0 (0%)	3 (7%)	NS
Cefoxitin	33 (89.2%)	2 (5.4%)	2 (5.4%)	37 (86%)	0 (0%)	6 (14%)	NS
Cefotaxime	34 (91.9%)	1 (2.7%)	2 (5.4%)	40 (93%)	0 (0%)	3 (7%)	NS
Cefepime	35 (94.6%)	0 (0%)	2 (5.4%)	36 (83.7%)	1 (2.3%)	6 (14%)	NS
Imipenem	12 (32.4%)	0 (0%)	25 (67.6%)	8 (18 .7%)	0 (0%)	35 (81.3%)	NS
Meropenem	13 (35.1%)	0 (0%)	24 (64.9%)	10 (23.2%)	0 (0%)	33 (76.8%)	NS
Gentamicin	29 (78.3%)	1 (2.7%)	7 (19%)	28 (65.1%)	2 (4.6%)	13 (30.3%)	NS
Tobramycin	27 (73%)	2 (5.4%)	8 (21.6%)	26 (60.4%)	0 (0%)	17 (39.6%)	NS
Amikacin	27 (73%)	2 (5.4%)	8 (21.6%)	25 (58.1%)	0 (0%)	18 (41.9%)	NS
Ciprofloxacin	26 (70.3%)	0 (0%)	11 (29.7%)	28 (65.1%)	0 (0%)	15 (34.9%)	NS
Levofloxacin	25(67.6%)	0 (0%)	12 (32.4%)	27 (62.7%)	0 (0%)	16 (37.3%)	NS
Ofloxacin	26 (70.3%)	0 (0%)	11 (29.7%)	28 (65.1%)	0 (0%)	15 (34.9%)	NS
Cefazoline	32 (86.5%)	0 (0%)	5 (13.5%)	40 (93%)	0 (0%)	3 (7%)	NS
Trimethprim-Sulfamethoxazole	30 (81.1%)	0 (0%)	7 (18.9%)	39 (90.6%)	0 (0%)	4 (9.4%)	NS
Nitrofurantoin (Additionaly for urine specimens)	12 (80%)	0 (0%)	3 (20%)	8 (72.7)	0 (0%)	3 (27.3%)	NS

Table 3. Biofilm formation capacity of *Kp* clinical isolates.

Characteristics	**hmv*Kp***	**c*Kp***	**X^2**	***P* value**
Biofilm producer	(29)78.4%	23 (53.5%)	5.4	0.01
Non biofilm producer	8 (21.6)	20 (46.5%)		
Total	37 (100%)	43 (100%)		

Chi-squared (χ2) test was used for statistical analysis. *P*-value > 0.05: Non-significant; *P*-value < 0.05: Significant; P-value < 0.01: Highly significant.

Table 4. Distribution of *rmpA* gene among hmv*Kp* and c*Kp* phenotypes by PCR assay.

	hmv*Kp* **(n= 37)**				**c*Kp*** **(n= 43)**				**X^2**	***P* value**
***rmpA* gene**	**Positive**		**Negative**		**Positive**		**Negative**		41.8	0.00001
	No.	**%**	**No.**	**%**	**No.**	**%**	**No.**	**%**		
	31	83.8	6	16.2	5	11.6	38	88.4		

Chi-squared (χ2) test was used for statistical analysis. *P*-value > 0.05: Non-significant; *P*-value < 0.05: Significant; *P*-value < 0.01: Highly significant.

Table 5. Association between biofilm formation and *rmpA* gene among hmv*Kp*.

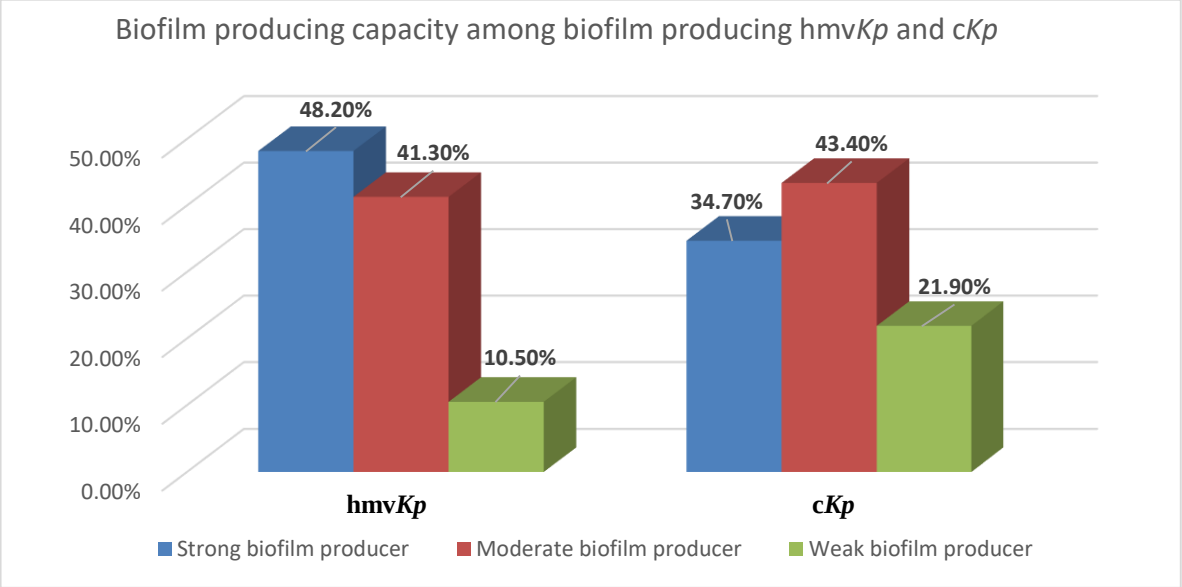
	Biofilm producers hmv*Kp* **(n= 29)**				**Non biofilm producers hmv*Kp*** **(n= 8)**				**X^2**	***P* value**
***rmpA* gene**	**Positive**		**Negative**		**Positive**		**Negative**		1.52	**0.217**
	No.	**%**	**No.**	**%**	**No.**	**%**	**No.**	**%**		
	24	82.7	5	7.3	5	62.5	3	37.5		

Chi-squared (χ2) test was used for statistical analysis. *P*-value > 0.05: Non-significant; *P*-value < 0.05: Significant; *P*-value < 0.01: Highly significant.

Table 6. Comparing string test results with rmpA gene results.

		***rmpA* gene**		
		Positive No. (36)	**Negative No. (44)**	
String test	**Positive No. (37)**	31(83.8%)	6 (16.2%)	PPV 83.78%
	Negative No. (43)	5 (11.6%)	38 (88.4%)	NPV 88.37%
		Sensitivity 86.11%	Specificity 86.36%	

Fig.4. Biofilm producing capacity among biofilm producing hmv*Kp* and c*Kp*.



Discussion

A new phenotype of called hv*Kp* was emerging in the last decades and was linked to life-threatening infections in young healthy and immunocompetent individuals [14].

The present study included 80 *K. pneumoniae* isolates, majority of isolates were from sputum 30 (37.5%) followed by urine 26 (32.5%) then pus 24 (30%). These results go in accordance with a study performed by **Nirwati et al.** and **Wang et al.** [15,16] and the Egyptian study by **Elsaid et al.** [17] as they reported that most of *K. pneumoniae* isolates were from respiratory specimens. In comparison to **Seifi et al.** [18] who found that most of *K. pneumoniae* were isolated from urine samples. Another study that performed in Egypt by **Elbrolosy et al.** [19] who reported the highest rate of *K. pneumoniae* was isolated from blood samples followed by respiratory secretions.

The present study reported that out of 80 *K. pneumoniae* isolates, 37(46.25 %) were identified by positive string test as hmv*Kp* while, the remaining 43 (53.75 %) isolates were string negative and classified as c*Kp*. This result agreed to a study performed in Egypt by **Abd-Elmonsef et al.** [20] who stated that the hypermucoviscous phenotype was identified in about 40% of all *K. pneumoniae* isolates. Also, **Jung et al.** [21] found in his study that 14 out of 33 (42.2%) were positive hv*kp* phenotype.

On the other hand, a study performed by **Alharbi et al.** [14] in Saudi Arabia reported a lower percentage of the hypermucoviscous phenotype (22.5% of all *K. pneumoniae* strains). While another study conducted in Iran by **Zamani et al.** [22] reported a higher percentage of hmv*Kp* (60.95%). The discrepancy of results may be due to the variance in sample size and geographical distribution.

The present study reported no statistically significant difference between hmv*Kp* and c*Kp* regarding the pattern of antimicrobial resistance with higher resistance to all beta-lactam antibiotics and cephalosporins but higher sensitivity towards imipenem and meropenem antibiotics. These results agree with **El-Mahdy et al.** [23] who found no significant difference between hmv*Kp* and c*Kp* strains in the resistance pattern. But **Alharbi et al.** [14] reported in their study that hmvKp strains was significantly lower resistance than in c*Kp* strains also, a study conducted in China by **Liu et al.** [24] reported the same results. In a study performed by **Imtiaz et al.** [4] in Pakistan, they stated higher resistance to cephalosporins, aminoglycosides, and fluoroquinolones but disagree regarding carbapenems resistance pattern. This may be attributed to the hyper-expression of the capsule that could provide a physical barrier against acquiring resistance plasmids [25].

Although different studies reported less resistance pattern in hmv*Kp*, a study performed by **Cubero et al.** [26] who Found that higher

antimicrobial resistance was found among hmv*Kp* that was acquired in hospitals . It might be due to misuse of antibiotics inside hospital settings that encourage the selection pressure for antimicrobial resistance.

The present study evaluated the biofilm formation and found that among the tested 37 hmv*kp* isolates, there were 29 (78.4.%) isolates as biofilm producer and 8 (21.6%) isolates were non biofilm producers. but for c*Kp* isolates, among the tested 43 isolates, there were 23 (53.5%) isolates as biofilm producer and 20 (46.5%) isolates were non biofilm producers. There was statistically significant difference regarding biofilm formation in hmv*Kp* and c*Kp* isolates. These results in concordance with **Alharbi et al.** and **Shah et al.** [14, 27] who reported that hmv*Kp* isolates were highly associated with biofilm formation. An important observation by **Wu et al.** [28] was that hmv*Kp* strains produced more biofilm than c*Kp* isolates, thereby suggesting that biofilm formation may be a influencing factor to its increased virulence. On the contrary, in the Egyptian study done by **El-Mahdy et al.** [23] There was no significant difference in biofilm production between hmv*Kp* in comparison to c*Kp*. This variation in results may be due to difference in sample size as **El-Mahdy et al.** reported only 13.8% hmv*Kp* isolates in their study.

The presence of *rmpA* gene is linked to hmvKp strains as it regulates the synthesis of extracellular polysaccharide capsule. This virulent phenotype is increasingly spreading as a cause of healthcare health associated infections. Fatal drawbacks were reported from infections caused by this virulent phenotype. So an efficient approach in preventing the increase in such infections could be done by active screening of *rmpA* gene in *Kp* isolates in healthcare settings [29].

The present study stated a highly statistically significant difference in the distribution of r*mpA* gene among hmv*Kp* and c*Kp* phenotypes. Among the hmv*Kp* clinical isolates, 31(83.8%) were harbouring the gene while 6 (16.2%) were lacking the gene while among c*Kp* clinical isolates, 5(11.6%) were harbouring the gene while it was absent in 38 (88.4%) of isolates. These results in agreement with jung et al., [21] who reported that up to 100% prevalence of rmpA in hypermucoviscous isolates. Also, **Guo et al.** and **Lin et al.** [30, 31] stated that *rmpA* gene was detected in 85.7%, 80.7% of strains of hv*Kp* respectively. In an Egyptian study by **Elbrolosy A et al.** [19] reported that the prevalence of associated *rmpA* gene was strongly associated with hmv*Kp* (80.8%) than c*Kp* strains (4.2%).

A lower percentage was reported in a study by **Imtiaz et al.** [4] who found that 13% of the MDR isolates were carried *rmpA* which is the first report for the presence of the *rmpA* gene in hv*Kp* isolates, with resistance to frontline antibiotics, from Pakistan. although the endemicity of hv*kp* in Asian region and also, they stated that 22% of the isolates exhibited a positive string test while only 11% of the isolates carried the *rmpA* gene. Another study done by **Qu et al.** [32] stated that 41% *rmpA* carriage in non-hypermucoviscous strains indicating additional underlying factors required for the hv*Kp* phenotype.

The current study assessed the accuracy of string test as a rapid method for detection of hmv*Kp*. On comparing the string test results using PCR as a gold standard for hypermucoviscosity, the sensitivity, PPV and NPV were evaluated. We found that the specificity was 86.36% and sensitivity was 86.11%. This result in concordance with **Russo et al. [**29] who reported results near to ours as they stated that string test had 91% sensitivity, 89% specificity. But **Elbrolosy et al.** and **Tan et al.** [19, 33] reported higher results regarding sensitivity 95.2%, 90.5% and lower regarding specificity 79.3%, 63.9% correspondingly.

The discrepancy in the results reported by many studies regarding the sensitivity and specificity of string test may be attributed to the difference in sample size and presence of several virulence genes including other regulator of mucoid phenotype A (*rmpA2*) and hypermucoviscosity-associated gene A (*magA*) that have been contribute to the hypervirulent phenotype.

Also, using the string test for defining hmvKp may be inadequately precise. Due to the presence of other underlining different virulence factors other than hypermucoviscosity that could contribute substantially to the virulence of hv*Kp* isolates [23].

Conclusion

Hmv*Kp* is being increased among *Kp* clinical isolates with high rate of biofilm formation that may lead to antimicrobial resistance among these strains. The string test is a simple and rapid screening test of hmv*Kp* that compared with detection of *rmpA* gene as a confirmatory tool for hypermucoviscosity.

Conflicts of interest

Authors have declared no conflicts of interest.

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Non delacred

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