Prevalence of plasmid-mediated resistance genes among multidrug-resistant uropathogens in Egypt

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Abstract

Background: The emergence of multidrug-resistant (MDR) uropathogens has become a public health threat and current knowledge of the genotypic basis of bacterial resistance is essential for selecting appropriate treatment options.

Objectives: To determine the prevalence of antimicrobial resistance among MDR uropathogens and to elucidate the molecular bases of plasmid-mediated resistance.

Methods: Bacterial isolates were recovered from urine specimens of 150 out-patients with signs and symptoms of urinary tract infections (UTIs) at El-Demerdash Hospital, Cairo, Egypt. Standard methods were used for identification, antimicrobial susceptibility testing was performed according to CLSI guidelines.

Results: Among the recovered isolates, 22.7% and 77.3% were Gram-positive, and negative, respectively. Of which; 43.3% were MDR with 60% harboring plasmids. Extended spectrum β-lactamase (ESBL) genes ^{bla}CTX-M, ^{bla}SHV, and ^{bla}TEM were detected on plasmids of 89.7%, 41%, and 84.6% of the tested isolates, respectively. The aminoglycoside resistance gene *aac6'-Ib/aac-6'-Ib-cr* was found on plasmids of 92.3% of the tested isolates followed by *qnrS* (92.3%), *qnrB* (46.2%), and *qnrA* (7.7%). The most prevalent quinolone efflux pump gene was *oqxB* (38.5%), followed by *oqxA* (20.5%), then *qepA* (10.3%).

Conclusion: High levels of resistance to nitrofurans, β-lactam/β-lactamase inhibitor, cephalosporins, aminoglycosides, and fluoroquinolones were detected, and their use as empirical treatment for UTIs has become questionable.

Keywords: ESBLs, *qnR*, plasmid-mediated, uropathgens, antibacterial resistance.

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Introduction

Urinary tract infection (UTI) is one of the most common infections worldwide which may be caused by Gram-negative or Gram-positive bacteria, as well as by some fungi. The most common causative organism is *Escherichia E. coli*[1]. Antimicrobial resistance (AMR) has become a major threat to public health in many countries. There has been a steady increase in AMR to the agents commonly used in treatment of UTIs[2,3]. In most UTI cases, empirical therapy is initiated before the results of urine culture and sensitivity are available, thus it is necessary to have AMR surveillance[4]. The emergence and spread of multidrug-resistant (MDR) organisms, which show resistance to three or more classes of antimicrobials[5], is increasing over time; and UTI cases requiring intravenous therapy due to the lack of effective oral treatment has become a challenge for physicians, complicating a previously simple-to-treat infection[3].

Extended Spectrum Beta Lactamases (ESBLs) have emerged as a chief mechanism of resistance among uropathogens[6]. These ESBLs are enzymes that trigger the resistance against β-lactam antibiotics by hydrolysis of the

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β-lactam ring[7]. Regrettably, ESBL-producing organisms usually carry resistance determinants to other antimicrobial agents as well,such as aminoglycosides and fluoroquinolones, leaving a limited range of treatment options[6]. The aim of this study was to reveal the prevalence of antimicrobial resistance and the molecular bases of plasmid-mediated resistance among bacterial uropathogens in one of the major clinical settings in Cairo, Egypt.

Materials and methods

Specimen collection

Starting October 2015 to May 2016, a total of 150 bacterial isolates were recovered from urine specimens of patients suffering signs and symptoms of UTIs at the outpatient clinics of El-Demerdash Hospital, Cairo, Egypt. All specimens were mid-stream urine and patients were instructed on how to collect specimens to avoid contamination. Patients included in the study were adults (ages ranging from 25 – 45 years), symptomatic, with pyuria (Pus cells ≥20/HPF), and the bacterial count in urine was $>10^5$ cfu/ml. The study was approved by Faculty of Pharmacy, Ain Shams University Ethics Committee Nr. 212 and an informed consent was obtained from patients after explaining the study purpose.

Identification of the recovered bacterial isolates

Isolates were categorized based on their Gram reactions, followed by identification using standard methods. Identification to the species level was done for MDR isolates by using API® 20E identification kit and API® Staph identification kit (BioMérieux, France) for Gram-negative and Gram-positive isolates respectively.

Antimicrobial susceptibility testing

The Kirby-Bauer disk diffusion test was performed according to Clinical and Laboratory Standards Institute (CLSI) guidelines[8,9] using commercially available antimicrobial disks (Oxoid, UK). The reference strains *E. coli* ATCC® 25922 and *Staphylococcus S. aureus* ATCC® 25923 were used for quality control. All MDR isolates were selected for further studying.

Determination of minimum inhibitory concentration of multidrug-resistant isolates

Minimum inhibitory concentration (MIC) values of MDR isolates were determined by broth microdilution method according to CLSI guidelines[9,10] using ceftriaxone, cefepime, meropenem, gentamicin, and ciprofloxacin. The reference strains *E. coli* ATCC® 25922 and *S. aureus* ATCC® 25923 were used for quality control.

Extraction of DNA plasmids from multidrug-resistant isolates

The extraction of DNA plasmids from MDR isolates was done using Zyppy™ Plasmid Miniprep Kit (Zymo Research, USA) according to the manufacturer's instructions. The extracted DNA plasmids were analyzed via agarose gel electrophoresis[11] and visualized by UV transilluminator.

Amplification of plasmid-encoded resistance genes

Amplification of antibiotic resistance genes was carried out by polymerase chain reaction (PCR) using the appropriate primers (Table 1); and the DNA plasmids of the MDR isolates as templates. Primers were manufactured by LGC Biosearch Technologies, USA. The amplified products were analyzed via agarose gel electrophoresis, and the expected DNA product size was determined by comparing to a 100 bp DNA ladder (New England Biolabs, UK).

The antibiotic resistance genes amplified in this study included ESBL genes (^{bla}CTX-M, ^{bla}SHV, and ^{bla}TEM); the *aac(6')-Ib* gene conferring resistance to aminoglycosides, and its bifunctional variant *aac(6')Ib-cr* conferring resistance to both aminoglycosides and ciprofloxacin; low level resistance plasmid-mediated quinolone resistance (PMQR) genes (*qnrA, qnrB, qnrS*), and quinolone efflux pump genes (*qepA, oqxA , oqxB*).

Sequencing of selected PCR products

Some selected PCR products of amplified genes were sent for sequencing at GATC, Germany using ABI 3730 xl DNA Sequencer. The alignment and assembly of the obtained forward and reverse sequence files into the final consensus was done using BioEdit v7.2.5 software[16].

Transformation

Plasmids extracted from the MDR isolates were used to transform competent *E. coli* DH5α prepared according to the modified Hanahan method[17] to test the phenotypic resistance of the transformants. Transformants were cultured on LB/ampicillin, LB/gentamicin, and LB/ciprofloxacin agar plates at concentrations of 100 μg/ml, 25 μg/ml, and 50 μg/ml, respectively.

Statistical analysis

Categorical variables were analyzed using the Chi-square test to determine statistical significance. Statistical analysis including descriptive statistics, frequency tables, and cross-tabulations was performed using Statistical Package for the Social Sciences software IBM® SPSS® version 20[18]. A value of $P<0.05$ was considered statistically significant, and significance was two-sided.

Results

Identification of the recovered bacterial isolates

Of the 150 recovered isolates; 79 (52.7%) were from female patients, and 71 (47.3%) were from male patients. Gram-negative bacilli (GNB) represented 77.3% of the total isolates (n=116); 107 of which (92.2%) were identified as members of *Enterobacteriaceae*, 7 (6%) were *Pseudomonas* spp., and 2 (1.7%) were *Acinetobacter* spp. On the other hand, 34 isolates (22.7%) were Gram-positive cocci (GPC); 23 of which (67.6%) were coagulase-negative *Staphylococcus* spp., 6 (17.6%) were *Staphylococcus aureus*, and 5 (14.7%) were *Enterococcus* spp.

Antimicrobial susceptibility testing

The antimicrobial susceptibility patterns of the recovered isolates are shown in Table 2. Out of 150 isolates; 65 (43.3%) were MDR; 51 of which (78.5%) were Gram-negative including *E. coli* (24/51; 47.1%), *Klebsiella* (*K.*) *pneumoniae* (15/51; 29.4%), *K. terrigena* (4/51; 7.8%), *Proteus mirabilis* (5/51; 9.8%), *Acinetobacter* (*A.*) *baumannii* (2/51; 3.9%), and *Pseudomonas* (*P.*) *aeruginosa* (1/51; 2%). Meanwhile; 14 (21.5%) of the 65 MDR isolates were Gram-positive including *S. aureus* (4/14; 28.6%), *S. haemolyticus* (3/14; 21.4%), *S. xylosus* (3/14; 21.4%), *S. epidermidis* (2/14; 14.3%), and *S. lentus* (2; 14.3%). Results of the antimicrobial suscepibility of the recovered isolates are presented in table S1. The prevalence of antimicrobial resistance of the tested MDR Gram-negative and Gram-positive isolates to different anti-microbial agents is shown in Figures 1 and 2, respectively.

Table 1. Primers sequences, expected product sizes, and annealing temperatures (T_a) of the tested genes

Gene	Primer	Primer sequence (5' → 3')	Expected product size (bp)	T_a (°C)	References
bla$_{CTX-M}$	P_f	CGCTTTGCGATGTGCAG	550	52	Bonnet *et al.*[12]
	P_r	ACCGCGATATCGTTGGT			
bla$_{SHV}$	P_f	GGTTATGCGTTATATTCGCC	867	52	Rasheed *et al.*[13]
	P_r	TTAGCGTTGCCAGTGCTC			
bla$_{TEM}$	P_f	ATGAGTATTCAACATTTCCG	867	50	
	P_r	CTGACAGTTACCAATGCTTA			
aac(6')-Ib /aac(6')Ib-cr	P_f	TTGCGATGCTCTATGAGTGG	358	46	Hamed *et al.*[14]
	P_r	CGTTTGGATCTTGGTGACCT			
qnrA	P_f	GCCCGCTTCTACAATCAAGT	347	60	Hamed *et al.*[15]
	P_r	GGCAGCACTATTACTCCCAAG			
qnrB	P_f	TATGGCTCTGGCACTCGTT	192	60	
	P_r	GCATCTTTCAGCATCGCAC			
qnrS	P_f	TCGGCACCACAACTTTTCAC	255	60	
	P_r	TCAC`ACGCACGGAACTCTAT			
qepA	P_f	TCTACGGGCTCAAGCAGTTG	312	55	
	P_r	ACAGCGAACCGATGACGAAG			
oqxA	P_f	CTCTCCTTTCTGCTCGTCGG	489	67	
	P_r	AATAGGGGCGGTCACTTTGG			
oqxB	P_f	TAGTGCTGGTGGTGCTGGTA	480	68	
	P_r	GGGTAGGGAGGTCTTTCTTCG			

Notes: *bla*$_{CTX-M}$, *bla*$_{SHV}$, and *bla*$_{TEM}$ genes code for ESBLs; *aac(6')-Ib* gene codes for aminoglycoside 6'-N-acetyltransferase type Ib; *aac(6')-Ib-cr* gene codes for aminoglycoside 6'-N-acetyltransferase type Ib ciprofloxacin-resistant variant; *qnrA, qnrB*, and *qnrS* genes are PMQR determinants coding for quinolone resistance; *qep A, oqxA*, and *oqxB* genes code for plasmid-mediated quinolone efflux pump resistance.

Abbreviations: T_a, calculated annealing temperature; ESBLs, extended-spectrum beta-lactamases; PMQR, plasmid-mediated quinolone resistance.

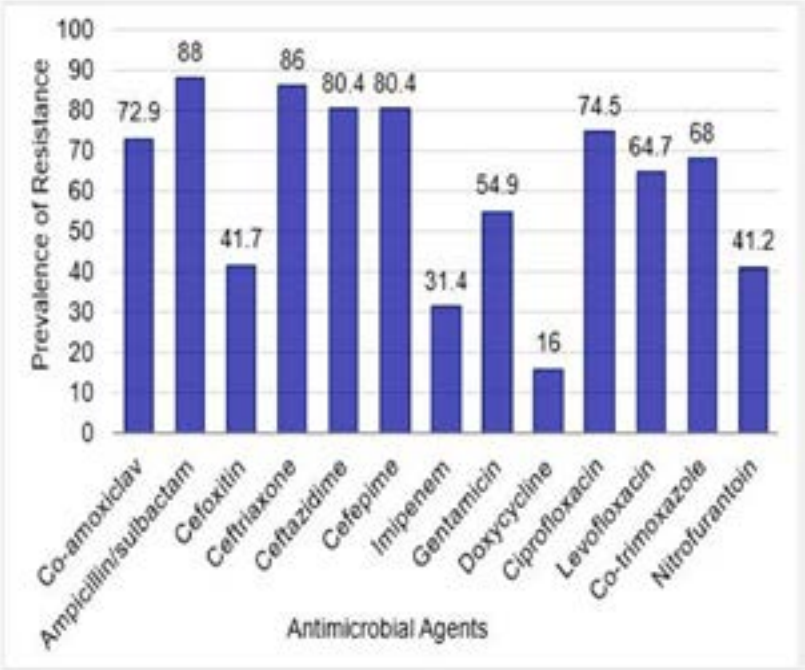

Figure 1. Prevalence of antimicrobial resistance of the 51 tested MDR GNB isolates to Different antimicrobial agents. Prevalence was expressed as percent of resistant isolates relative to total tested isolates for each antimicrobial agent

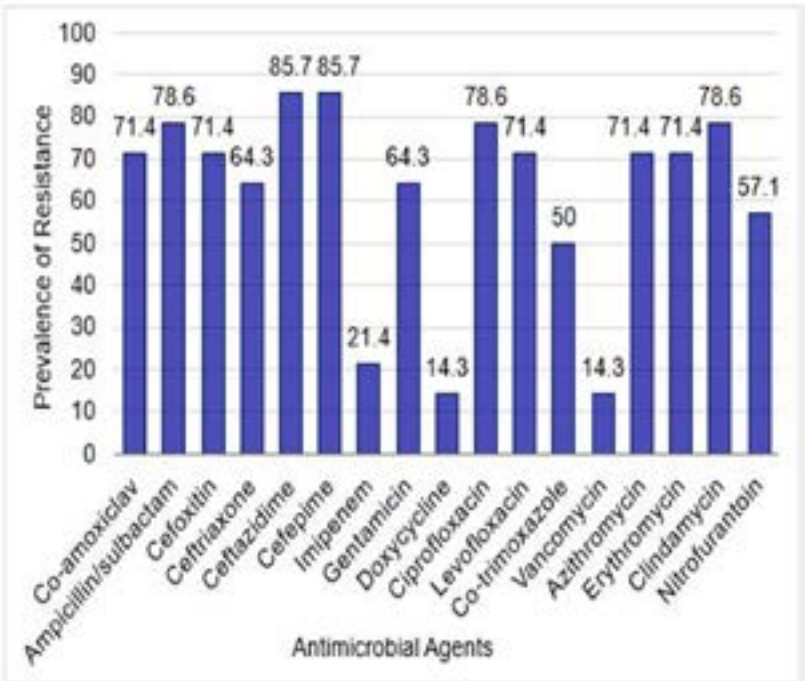

Figure 2. Prevalence of antimicrobial resistance of the 14 tested MDR GPC isolates to different anti-microbial agents. Prevalence was expressed as percent of resistant isolates relative to total tested isolates for each antimicrobial agent

Minimum inhibitory concentrations of MDR isolates

The obtained MIC results of MDR GNB and GPC isolates are shown in supplementary tables S2 and S3, respectively.

Table 2. Antimicrobial susceptibility patterns of the recovered 150 isolates

Antibiotics	Resistant isolates, n (%)					
	Enterobacteriaceae (n=107)	*Pseudomonas* spp. (n=7)	*Acinetobacter* spp. (n=2)	*Staphylococcus aureus* (n=6)	Coagulase -negative *Staphyloc occus* spp. (n=23)	*Enterococcus* spp. (n=5)
	R	R	R	R	R	R
Co-amoxiclav	53 (49.5)	nd	nd	2 (33.3)	9 (39.1)	nd
Ampicillin/ sulbactam	55 (51.4)	nd	2 (100)	3 (50.0)	9 (39.1)	nd
Cefoxitin	24 (22.4)	nd	nd	3 (50.0)	10 (43.5)	nd
Ceftriaxone	42 (39.3)	nd	1 (50.0)	1 (16.7)	8 (34.8)	nd
Ceftazidime	44 (41.1)	3 (42.9)	2 (100)	3 (50.0)	10 (43.5)	nd
Cefepime	46 (43.0)	1 (14.3)	1 (50.0)	3 (50.0)	9 (39.1)	nd
Imipenem	17 (15.9)	0 (0.0)	2 (100)	1 (16.7)	2 (8.7)	nd
Gentamicin	26 (24.3)	1 (14.3)	2 (100)	1 (16.7)	8 (34.8)	nd
Doxycycline	12 (11.2)	nd	0 (0.0)	1 (16.7)	1 (4.3)	0 (0.0)
Ciprofloxacin	43 (40.2)	2 (28.6)	2 (100)	1 (16.7)	11 (47.8)	1 (20.0)
Levofloxacin	35 (32.7)	2 (28.6)	1 (50.0)	2 (33.3)	8 (34.8)	1 (20.0)
Co-trimoxazole	40 (37.4)	nd	2 (100)	2 (33.3)	6 (26.1)	nd
Vancomycin	nd	nd	nd	1 (16.7)	1 (4.3)	1 (20.0)
Azithromycin	nd	nd	nd	3 (50.0)	8 (34.8)	nd
Erythromycin	nd	nd	nd	3 (50.0)	8 (34.8)	2 (40.0)
Clindamycin	nd	nd	nd	3 (50.0)	9 (39.1)	nd
Nitrofurantoin	40 (37.4)	nd	nd	6 (100)	11 (47.8)	0 (0.0)

Abbreviations: R, resistant; nd, not determined (due to lack of interpretation data in CLSI guidelines).

Table 3. Percentage of plasmid-mediated antimicrobial resistance genotypes of MDR isolates and statistically significant associations

MDR isolates harboring plasmids	Genotypes	No. of isolates (%*)	Significant associations	Pearson chi-square
Positive (n=39)	*bla_{CTX-M}/bla_{TEM}/aac(6')-Ib/qnrS*	5 (7.7%)	[a]co-amoxiclav/*bla_{SHV}*	0.026
	bla_{CTX-M}/bla_{TEM}/aac(6')-Ib/qnrS/oqxB	4 (6.2%)	[a]cefoxitin/*bla_{SHV}*	0.002
	bla_{CTX-M}/bla_{SHV}/bla_{TEM}/aac(6')-Ib/qnrB/qnrS	4 (6.2%)	[a]imipenem/*bla_{SHV}*	0.012
	bla_{CTX-M}/bla_{SHV}/aac(6')-Ib/qnrB/qnrS	3 (4.6%)	[a]meropenem/*bla_{SHV}*	0.01
	bla_{CTX-M}/bla_{SHV}/bla_{TEM}/aac(6')-Ib/qnrS	2 (3.1%)		
	bla_{CTX-M}/bla_{TEM}/aac(6')-Ib/qnrS/oqxA/oqxB	2 (3.1%)	[b]*bla_{CTX-M}/aac(6')-Ib*	0.001
	bla_{CTX-M}/aac(6')-Ib/qnrB/qnrS	2 (3.1%)	[b]*bla_{SHV}/qnrB*	0.018
	bla_{CTX-M}/bla_{TEM}/aac(6')-Ib/qnrB/qnrS	2 (3.1%)	[b]*bla_{SHV}/qnrS*	0.031
	bla_{CTX-M}/bla_{SHV}/bla_{TEM}/aac(6')-Ib/qnrB	2 (3.1%)	[b]*bla_{SHV}/oqxB*	0.035
	bla_{SHV}/bla_{TEM}	1 (1.5%)	[b]*bla_{TEM}/qnrB*	0.047
	bla_{CTX-M}/bla_{TEM}/aac(6')-Ib/qnrB/qnrS/oqxA/oqxB	1 (1.5%)	[b]*qnrB/oqxB*	0.01
	bla_{CTX-M}/bla_{TEM}/aac(6')-Ib/qnrA/qnrS/oqxB	1 (1.5%)		
	bla_{CTX-M}/bla_{SHV}/bla_{TEM}/aac(6')-Ib/qnrS/oqxA/oqxB/qepA	1 (1.5%)		
	bla_{TEM}/qnrB/qnrS	1 (1.5%)		
	*bla_{CTX-M}/bla_{SHV}/bla_{TEM}/aac(6')-Ib/qnrB/qnrS/oqxA/oqxB/*qepA	1 (1.5%)		
	bla_{SHV}/bla_{TEM}/aac(6')-Ib/qnrB/qnrS	1 (1.5%)		
	bla_{CTX-M}/qnrS/oqxA/oqxB	1 (1.5%)		
	bla_{CTX-M}/bla_{TEM}/aac(6')-Ib/qnrA/qnrS/oqxB/qepA	1 (1.5%)		
	bla_{CTX-M}/bla_{SHV}/bla_{TEM}/aac(6')-Ib/qnrS/oqxA/oqxB	1 (1.5%)		
	bla_{CTX-M}/bla_{TEM}/aac(6')-Ib/qnrA/qnrS	1 (1.5%)		
	bla_{CTX-M}/bla_{TEM}/aac(6')-Ib/qnrB/qnrS/oqxA/oqxB/qepA	1 (1.5%)		
	bla_{TEM}/aac(6')-Ib/qnrS/oqxB	1 (1.5%)		
Negative (n=26)	---	26 (40%)		

Notes: genotypes, plasmid-mediated antimicrobial resistance. *Percentages were calculated with reference to the number of MDR isolates (n=65). [a]Significant association between antibiotic resistance and PCR detection of the respective gene on plasmids. [b]Significant co-existence of resistance genes on plasmids of the same isolate.
Abbreviations: MDR, multidrug-resistant; PCR, polymerase chain reaction.

Extraction of DNA Plasmids from MDR isolates

DNA plasmids were extracted from 39 (60%) of the 65 MDR isolates. The extracted plasmids were analyzed via agarose gel electrophoresis, and the band sizes were compared to a 1 kb DNA ladder (New England Biolabs, UK).

Amplification of some plasmid-encoded resistance genes

Results of PCR amplification of the ESBL genes (^{bla}C-TX-M, ^{bla}SHV, and ^{bla}TEM); the *aac(6')-Ib* gene, *plasmid*-mediated quinolone resistance (PMQR) genes (*qnrA, qnrB, qnrS*), and quinolone efflux pump genes (*qepA, oqxA , oqxB*) are depicted in figures S1, S2 and S3. The prevalence of amplified antibiotic resistance genes among MDR bacterial uropathogens is shown in Figure 3.

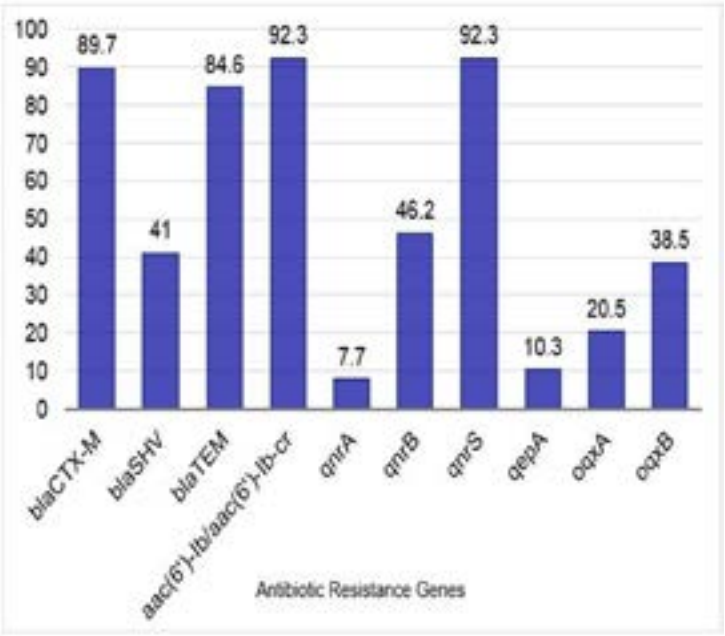

Figure 3. Prevalence of some selected antibiotic resistance genes among MDR bacterial uropathogens. Prevalence was expressed as percent of isolates carrying the tested genes relative to total tested isolates (n=39)

Genotypes of MDR isolates

Among the 65 MDR isolates; 22 different genotypes were observed based on the PCR detection of antimicrobial resistance genes on the extracted DNA plasmids of the MDR isolates, as shown in Table 3.

Transformation

The results of transformation along with PCR amplification for the tested isolates are shown in Table S4. In case of plasmids harboring ESBL coding genes (n=39), successful transformation and gene expression was achieved with plasmids extracted from 28 isolates (71.8%) harboring such plasmids; demonstrated by the ability of transformants to grow on LB/ampicillin agar plates. Out of 36 MDR isolates that carried the aminoglycoside resistance gene *aac(6')-Ib/aac(6')-Ib-cr*, the plasmids extracted from 15 isolates (41.7%) were successfully transformed and resistance genes were expressed; which was demonstrated by the ability of transformants to grow on LB/gentamicin agar plates. Isolates that carried any of the plasmid-mediated quinolone resistance (PMQR) genes (*aac(6')-Ib-cr, qnrA, qnrB, qnrS, qepA, oqxA*, and *oqxB*) were found to be 38 out of the 39 tested MDR isolates (97.4%); plasmids extracted from 10 of which (26.3%) were successfully transformed and resistance genes were expressed; demonstrated by the ability of transformants to grow on LB/ciprofloxacin agar plates.

Statistical analysis

Statistical analysis has shown that there is significant association between antimicrobial resistance and PCR detection of the respective genes on DNA plasmids. There is also significant co-existence of PCR-detected antibiotic resistance genes on DNA plasmids of the same isolate (P value <0.05). The statistical association and respective P values are shown in Table 3.

Discussion

As reported, UTIs are becoming more difficult to treat due to the emergence and prevalence of a wide range

of antibiotic resistance mechanisms[3]. Accordingly, in this study we assessed both the phenotypic and genotypic bases of antimicrobial resistance of some MDR uropathogens against the most common antimicrobial agents used in treatment of UTIs.

The antimicrobial susceptibility of the GPC collected in this study (n=34) showed that the lowest resistance was observed to doxycycline, vancomycin, and imipenem (5.9%, 8.8%, 10.3%). On the other hand, the highest resistance was observed to nitrofurantoin (50.0%); cefoxitin, ceftazidime (44.8% each); ampicillin/sulbactam and cefepime (41.4% each). The antimicrobial susceptibility of the GNB (n=116) showed that the lowest resistance was observed to doxycycline (11.0%) and imipenem (16.4%). The highest resistance was observed to ampicillin/sulbactam (52.3%); co-amoxiclav (49.5%); ceftazidime (42.2%); cefepime (41.4%); and ciprofloxacin (40.5%). These results limit the use of nitrofurans, cephalosporins, β-lactam/β-lactamase inhibitors, and fluoroquinolones as empirical treatment of UTIs, while tetracyclines and carbapenems still retain their efficacy in treating UTIs based on *in vitro* data. Some other studies deduced that imipenem showed highest efficacy and may be the drug of choice for empirical therapy of UTIs based on the in vitro data[19-21].

In this study, PCR amplification was used to detect some plasmid-mediated antimicrobial resistance genes associated with the 39 MDR isolates harboring plasmids. The ESBLs genes were detected in all 39 (100%) isolates with *bla*CTX-M gene showing highest prevalence (89.7%), followed by *bla*TEM (84.6%) then *bla*SHV (41%). Our findings agreed with the study conducted by Goudarzi et al. in 2015 which also revealed a dominant presence of *bla*CTX-M (74.9%), followed by *bla*TEM (70%), then *bla*SHV (59.9%)[22]. Of the 39 MDR isolates harboring plasmids, 36 (92.3%) carried the *aac6'-Ib/aac-6'-Ib-cr* gene conferring resistance to aminoglycosides and ciprofloxacin, 11 of which (30.6%) carried the three ESBLs genes together, and 23 (63.9%) carried two of the ESBLs genes. A study conducted by Paterson *et al.* revealed that ESBL-producing *K. pneumoniae* isolates that were ciprofloxacin-resistant possessed multiple β-lactamases[23], to which our results are in accordance. Our results also showed that there is a significant association between the presence of *bla*CTX-M gene and *aac6'-Ib/aac-6'-Ib-cr* gene in the same isolate (P=0.001). Accordingly, isolates that produce ESBLs also carry resistance genes to aminoglycosides and fluoroquinolones, thus reducing treatment options. This cross-resistance is more prominent in urinary isolates[3]. Therefore, carbapenems, which are less prone to hydrolysis by such enzymes, have become the preferred therapy for infections with ESBL-producing pathogens[24–27].

The most prevalent *qnr* gene in our study was *qnrS* gene (36/39; 92.3%), followed by *qnrB* (18/39; 46.2%), and *qnrA* (3/39; 7.7%). This prevalence pattern is in accordance to that obtained in a study conducted by several recent studies[28-30]. The quinolone efflux pump resistance genes were detected in 15 (38.5%) out of the 39 MDR isolates and their prevalence was highest for *oqxB* (15/39; 38.5%), followed by *oqxA* (8/39; 20.5%), and *qepA* (4/39; 10.3%). It should be noted that the acquisition of PMQR genes alone results in low levels of resistance to fluoroquinolones, and does not cause MICs to exceed the breakpoints of these agents[31], but rather facilitates the selection of mutants of higher-level resistance[32]. This was evident in our study by the lack of statistically significant association between the presence of PMQR genes and resistance to fluoroquinolones, which means the presence of PMQR genes alone, did not confer resistance to fluoroquinolones.

Conclusion

High levels of resistance to antimicrobials commonly used for treatment of UTIs was detected among MDR uropathogens. The current efficacy of nitrofurans, β-lactam/β-lactamase inhibitor, cephalosporins, aminoglycosides, and fluoroquinolones has become questionable. Carbapenems, tetracyclines, and vancomycin have yet to retain their efficacy in treatment of UTIsbased on *in vitro* data. No significant correlation was observed between the presence of PMQR genes and fluoroquinolone resistance, indicating that PMQR genes alone do not grant phenotypic resistance to fluoroquinolones, however the resistance may have been due to co-existence of ESBL and/or *aac6'-Ib/aac-6'-Ib-cr* genes in the same isolate or even on the same plasmids.

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Conflict of Interest Disclosure

The authors declare that they have no conflict of interests

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