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RESISTOME AND PLASMIDOME IN AN EXTENSIVELY DRUG-RESISTANT EXTRA-INTESTINAL PATHOGENIC *ESCHERICHIA COLI* ISOLATED FROM A ONE-YEAR-OLD CHILD WITH RESPIRATORY DISTRESS SYNDROME

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SUMMARY

The global health is facing a growing menace of multidrug-resistant and extensively drug-resistant bacteria over time. Genomic plasticity of pathogenic *Escherichia coli* allows them to continuously develop and acquire genetic elements associated with antibiotic resistance, including resistance to last resort drugs like carbapenems – only used for the treatment of multidrug-resistant bacterial infections. This study aimed at identifying resistome and plasmidomes carrying antibiotic-resistant genes (ARGs) in an extensively drug-resistant extra-intestinal pathogenic *E. coli* strain XP817 isolated from a one-year-old child with respiratory distress syndrome. Whole genome analysis of strain XP817 revealed 68 ARGs documented in the Comprehensive Antibiotic Resistance Database (CARD). Notably, strain XP817 possessed 12 ARGs and mutations responsible for its extensively drug-resistant phenotype to aminoglycosides (*aac*C2 and *rmt*B), tetracycline (*tet*A and *tet*R), bleomycin (bla_{MBL}), chloramphenicol (*mdt*M), penicillin and cephalosporin ($bla_{CTX\text{-}M\text{-}27}$ and bla_{AmpC}), fluoroquinolones (*gyr*A and *par*C), macrolide (*mph*A) and carbapenem ($bla_{NDM\text{-}1}$). Furthermore, five plasmids carrying seven ARGs originated from *E. coli* strain ECCHD184 (carrying *mph(*A*)*) and other species of *Enterobacteriaceae* family including *Enterobacter hormaechei* (carrying $bla_{NDM\text{-}1}$ and bla_{MBL}), *Proteus mirabilis* (carrying *tet(*A*)* and *tet*R), *Raoultella ornithinolytica* (carrying $bla_{CTX\text{-}M\text{-}27}$) and *Klebsiella pneumonia* (carrying *aac*C2) were detected. In conclusion, our study underlines the crucial role of the ARG dissemination via horizontal gene transfer in *E. coli* as well as in the *Enterobacteriaceae* family.

Keywords: Extra-intestinal pathogenic Escherichia coli (ExPEC), resistome, plasmidome, extensively drug resistance, carbapenem resistance, whole-genome sequencing.

INTRODUCTION

The emergence of carbapenem-resistant *Enterobacteriaceae* (CRE) is threatening the global public health system. In 2017, World Health Organization (WHO) published a global priority list of pathogens in which CRE ranked in the highest priority category

(http://www.who.int/medicines/publications/WHO-PPL-Short_Summary_25Feb-ET_NM_WHO.pdf.). To address this global epidemic, surveillance of CRE is urgently needed in all healthcare settings. Notably, major mechanisms of carbapenem resistance are due to carbapenemase enzymes, which are encoded by genes *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{VIM}$, *bla*$_{IMP}$ or *bla*$_{OXA-48}$ located mainly on mobile elements and are highly transmissible (Cui, Zhang, & Du, 2019). In South-East Asia and Vietnam, the majority of CRE strains harbors *bla*$_{KPC}$ and *bla*$_{NDM-1}$, which emphasizes the regional dissemination of CRE depending on the horizontal transfer of their plasmid-mediated genes (Tran *et al.*, 2019; Tran et al., 2015; Zhang *et al.*, 2017). In 2019, massive surveillance in Vietnamese hospitals assumed that 52% of inpatients were colonized with CRE and the colonization rate drastically proportions with treatment day as well as mortalities caused by the hospital -acquired infections (Tran *et al.*, 2019). Another study spotted an arising tendency of New Delhi metallo-beta lactamase (NDM) - producing CRE that is an aggregation of *bla*$_{NDM-1}$ and relevant antibiotic-resistant genes (ARGs) against other antibiotic classes such as aminoglycosides, quinolones or fluoroquinolones on the same plasmid (Hirabayashi *et al.*, 2020). As a result, these specific organisms are believed to develop into various highly drug-resistant forms including multidrug-resistant (MDR), extensively drug-resistant (XDR) or pandrug-resistant (PDR) pathogens. Thus, identification of resistant factors and comprehending their acquisition and dissemination in the population are the basics to control better the emergence and transmission of highly drug-resistant strains in the healthcare settings as well as in the community.

Recognized as one of the most common infectious pathogens, extra-intestinal pathogenic *Escherichia coli* or ExPEC are responsible for a plethora of infections in both human and animals. ExPEC is comprised of pathotypes like uropathogenic *E. coli* (UPEC), avian pathogenic *E. coli* (APEC), meningitis and sepsis meningitis-associated *E. coli* (MNEC). The pathotypes have been transmitted globally and became reservoirs of MDR strains. In this study, we provide an in-depth characterization of the resistome and plasmidome of a XDR ExPEC strain which resists to carbapenems using a comparative genomic analysis.

MATERIALS AND METHODS

Clinical *E. coli* strains

The carbapenem-resistant *E. coli* XP817 was isolated from the pleural fluid of a one-year-old boy diagnosed with respiratory distress syndrome at Saint-Paul hospital in 2012. While carbapenem-sensitive *E. coli* TN1393 was isolated from the urine sample of a 28-year-old woman suffering from kidney infection in Thanh Nhan hospital in 2013. These strains were cultured onto blood agar medium at 37°C. The pure colonies were collected and identified as *E. coli* using MALDI-TOF MS system (Bruker, Germany). Bacterial suspensions in glycerol 50% were stored in -80°C for further use.

Antimicrobial susceptibility testing

The antimicrobial susceptibility of *E. coli* strains was conducted against 23 drugs categorized into 8 antibiotic groups including penicillins (ampicillin 10 µg, oxacillin 5 µg, ticarcillin 75 µg, and piperacillin 100 µg), cephalosporins (cephalexin 30 µg, cefoxitin 30 µg,

cefpopdoxim 10 µg, and cefepim 30 µg), carbapenems (ertapenem 10 µg, imipenem 10 µg, and meropenem 10 µg), monobactams (aztreonam 30 µg), β-lactam combination agents (amoxicillin-clavulanate 20/10 µg, piperacillin-tazobactam 100/10 µg, and ticarcillin-clavulanate 75/10 µg), aminoglycosides (amikacin 30 µg, gentamicin 10 µg, and tobramycin 10 µg), tetracycline (tetracycline 30 µg), quinolones (ciprofloxacin 5 µg, levofloxacin 5 µg, ofloxacin 5 µg, and nalidixic acid 30 µg) (SirScan/i2a Diagnostics, France). The suspension of *E. coli* strains was prepared on Muller-Hinton broth (MHB) to reach 0.5 McFarland turbidity standards. Next, each 100 µL of suspension was spread onto a Mueller-Hinton agar (MHA) plate to establish bacterial lawn and the antibiotic discs were placed on top. The inhibition zone diameter was measured after 24 h of incubation at 37°C. The experiment was repeated three times and the mean of the inhibition zone was used to interpret the antimicrobial susceptibility in accordance with Clinical and Laboratory Standards Institute M100-ED30:2020 (CLSI 2020). *E. coli* ATCC 25922 was used as a positive control for all experiments.

Minimum inhibitory concentration

The minimum inhibitory concentration (MIC) of carbapenems (imipenem and meropenem) against *E. coli* strains was determined by broth microdilution assay (CLSI 2020). The bacterial suspension was freshly prepared in Mueller Hilton broth (MHB) to obtain a final concentration of 10^8 CFU/ml. Then, 180 µl of the suspension culture was transferred into each 96-microplate well followed by adding 20 µl of tested antibiotic solutions separately to obtain final concentrations ranging from 0 to 128 µg/ml. The plates were incubated at 37°C, and the optical density (OD) was measured at 600 nm after 18-24 h of incubation using a microplate reader SpectraMax iD5 (Molecular Devices, USA). *E. coli* ATCC 25922 was used as a positive control. All experiments were performed in triplicate under the same conditions. The MIC value was determined as the lowest concentration showing 95% inhibition of bacterial growth.

Genomic DNA extraction, detection of carbapenem-resistant genes and whole-genome sequencing

Bacterial DNA was extracted with Norgen Bacterial Genomic DNA Isolation Kit (Norgen Biotek, Canada) following the manufacturer's instructions. The DNA purity and concentration was qualified by Nanodrop 2000 Spectrophotometer (Thermo Fisher Scientific, USA). Genomic DNA was then screened for carbapenem-resistant genes including *bla*KPC, *bla*NDM, *bla*IMP, *bla*OXA48, and *bla*VIM by real-time PCR assay as described previously (van der Zee *et al.*, 2014). For whole genome sequencing, the extracted DNA was used for library preparation and sequenced using an Illumina HiSeq 4000 system (Illumina, San Diego, CA, USA) at the Beijing Genomics Institute (Shenzhen, China).

***De novo* whole genome assembly**

FastQC *v0.11.9* was used to examine the quality of the bases, adaptor sequences and overrepresented sequences of the raw data sequences. Low-quality bases with a Phred quality score below 20 were depurated from both forward and reverse reads by Trimmomatic *v.0.39*. Trimmed reads were then assembled by SPAdes

v3.14.1. The genome was assembled by QUAST *v5.0.2*. All statistics of QUAST were based on contigs larger than 500 bp.

Phylogenetic tree analysis

Genomic sequences of twelve representative *E. coli* strains were retrieved from NCBI GenBank for phylogeny analysis. The phylogenetic trees were established based on two approaches, 16S ribosomal RNA (rRNA) and multi-locus sequence typing (MLST). The 16S rRNA sequences of strains *E. coli* XP817 and TN931 were extracted by Barrnap *v0.9* and seven housekeeping genes (*adk, fumC, gyrB, icd, mdh, purA* and *recA*) of each strain were detected by MLST 2.0. Phylogenetic trees were created on the phylogeny.fr platform via the following parameters: MUSCLE for multiple alignment, Gblocks for alignment curation, PhyML for construction of phylogenetic tree via maximum likelihood approach, and TreeDyn for tree visualization.

Detection of antibiotic resistome and plasmidomes

Detection of ARGs was carried out by using the RGI 5.1.1 (Resistance Gene Identifier) and CARD 3.1.0 (Comprehensive Antibiotic Resistance Database Resistance Gene Identifier) (Alcock *et al.,* 2020). Additionally, plasmid assemblies were constructed from the raw reads by plasmidSPAdes (Antipov *et al.,* 2016) and then compared to the non-redundant nucleotide database utilizing the online NCBI-BLASTn tool (https:/blast.ncbi.nlm.nih.gov/Blast.cgi) to classify and define contigs originated from plasmids. Lastly, Rapid Annotation using Subsystem Technology (RAST) annotation pipeline (Bankevich *et al.,* 2012) illustrated the genome features and the function of protein-encoding gene in the draft genomes and the predicted plasmids.

RESULTS AND DISCUSSION

Antibiotic-resistant phenotypic profile of *E. coli* XP817 and TN3193

Antibiotic-susceptibility testing results showed that *E. coli* strain XP817 was completely resistant to tested 22 antibiotics (Table 1), underlining the extensively drug-resistant phenotype. Whereas, *E. coli* strain TN3193 was sensitive to all tested antibiotics then was classified as pan-susceptible strain. The *E. coli* strain XP817 exhibited MICs of > 128 µg/ml for carbapenems (imipenem and meropenem), suggesting the treatment regimen is very complicated. Globally, in 2013, carbapenem-resistant *E. coli* was reported in 26 countries with the highest percentage reported in India (10.5%), followed by Vietnam (8.9%) and Bulgaria (3.4%) (Kelly *et al.*,2017). The recent national survey in 2017 showed the prevalence of CRE was 11%, suggesting that the continue increase and spread of CRE in the hospitals as well as in community (Vu *et al.,* 2021). A study from Vietnam showed that CRE infections are associated with the increase of mortality and health care costs (Tran *et al.,* 2019). Therefore, continue monitoring the emergence and spread of the CRE strains, are needed to reduce the impact of resistance.

Table 1. Antibiotic-resistant phenotypic profile of *E. coli* TN1393 and XP817.

Antibiotic Class	Antibiotic	TN1393		XP817	
		Inhibition zone diameter (mm)	AST result	Inhibition zone diameter (mm)	AST result
Penicillins	Ampicillin 10 µg	14.3	S	0	R
	Oxacillin 5 µg	21	S	0	R
	Piperacillin 100 µg	17.0	S	12.3	R
	Ticarcillin 75 µg	23.0	S	0	R
B-lactams combination	Amoxicillin-Clavulanate 20/10 µg	15.3	S	0	R
	Piperacillin-Tazobactam 100/10 µg	25	S	11.3	R
	Ticarcillin-Clavulanate 75/10 µg	20.7	S	8.7	R
Cephalosporins	Cephalexin 30 µg	20.0	S	0	R
	Cefoxitin 30 µg	24.0	S	0	R
	Cefpopdoxim proxetil 10 µg	22.0	S	0	R
	Cefepim 30 µg	25.7	S	14	R
Carbapenems	Etarpenem 10 µg	31.3	S	12.7	R
	Imipenem 10 µg	28.7	S	14.7	R
	Meropenem 10 µg	29.3	S	12.7	R
Monobactams	Aztreonam 30 µg	31	S	19	I
Tetracycline	Tetracycline 30 µg	19.7	S	0	R
Aminoglycosides	Amikacin 30 µg	16.7	S	0	R
	Gentamicin 10 µg	18.7	S	0	R
	Tobramycin 10 µg	14.7	S	0	R
Quinolones	Nalidixic acid 30 µg	22.3	S	0	R
	Ofloxacin 5 µg	24.0	S	0	R
	Ciprofloxacin 5 µg	6.3	S	0	R
	Levofloxacin 5 µg	29.0	S	0	R

R: resistant; I: intermediate-resistant; S: susceptible.

Identification of Carbapenem-resistant genes by real time PCR

Real time PCR screening of carbapenem-resistant genes detected the presence of *bla*NDM in *E. coli* strain XP817, demonstrating the high resistant level to carbapenems. A study reported for the first time a NDM-1-producing *E. coli* with the MIC of meropenem of >128 µg/ml isolated

from a patient admitted to a surgical hospital in Hanoi in 2010 (H. H. Tran et al., 2015). Subsequent studies in Vietnam have reported a range of carbapenem resistance genes in CRE including *bla*KPC, *bla*KPC-2, *bla*NDM-1, *bla*NDM-4, *bla*NDM-5, *bla*OXA and *bla*OXA-48 (Malchione *et al.*, 2019). In accordance to previous studies, the *E. coli* strain XP817 collected in our study is among the first CRE population detected in Vietnam. Despite the rapid dissemination of CRE in Vietnam, comprehensive molecular mechanisms associated with carbapenem resistance were not fully investigated. Therefore, it is urgently needed to adopt advanced whole genome sequencing technology for better understanding the evolution of antibiotic resistance mechanisms to control the emergence and transmission of CRE in hospitals and in the community.

De novo assembly of whole genome and phylogenetic analysis

The assembled genome of *E. coli* XP817 revealed a genome size of 5.2 Mb, consisting of 5284 coding sequences and GC content of 50.7%. The genome of *E. coli* TN1393 had a size of 5.7Mb, containing 5972 coding sequences and GC content of 50.3%. The 16S rRNA sequence-based phylogenetic tree showed that two isolates XP817 and TN1393 were clustered in two molecular clades on the same branch (Figure 1A). *E. coli* TN1393 and *E. coli* XP817 were assigned to ST73 and ST167, respectively, according to the MLST Pasteur scheme. Accordingly, these two strains were distributed on different phylogenetic clades (Figure 1B). The ST73 was claimed as a predominant clone affecting urinary tract infection (UTI) with a wide-range virulence gene-profile (de Souza da-Silva *et al.*, 2017), while the ST167 belonged to ST10 clonal complex is the common clone producing extended-spectrum beta-lactamase (ESBL) and carbapenemase (Zong, Fenn, Connor, Feng, & McNally, 2018). Notably, the ST617 was one of the four global carbapenemase-producing *E. coli* causing many hospital-acquired infection outbreaks (Peirano *et al.*, 2022). Because *E. coli* is mainly responsible for hospital-acquired and community-associated infections, *E. coli* is an important reservoir for the acquisition and dissemination of ARGs via horizontal gene transfer (HGT) mechanisms.

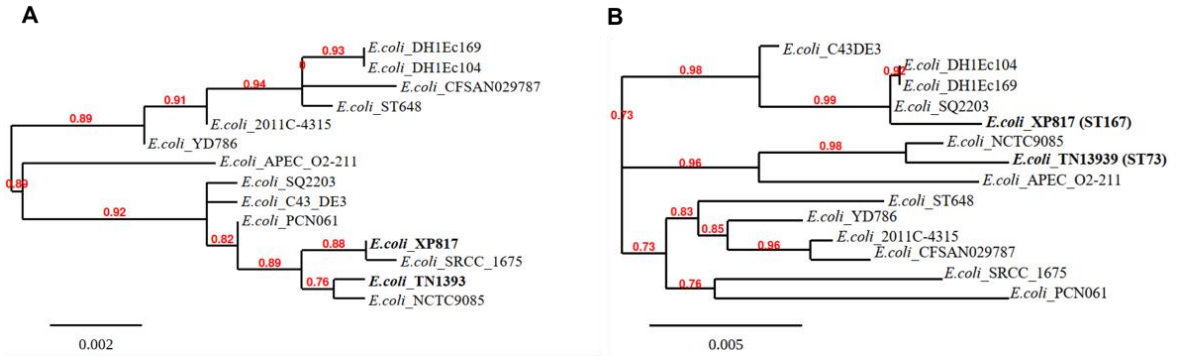

Figure 1. The phylogenetic trees based on 16S rRNA gene sequence (**A**) and MLST (**B**) of *E. coli* XP817 and TN1393.

Antibiotic resistome of XDR *E. coli* XP817

In total 68 ARGs were detected in the genome of *E. coli* XP817 by searching on the CARD-RGI database. The ARG annotation list in corresponding to different resistance mechanisms is summarized in Table 2. Among them, 12 ARGs (in bold, Table 2) conferring resistance to important antibiotics used clinically to treat Gram-negative infections demonstrating its XDR genotype. Specifically, the co-existence of *aac*C2 and *rmt*B genes encoding for aminoglycoside N(3)-acetyltransferase and 16S rRNA methyltransferase (RmtB) enzymes, respectively, is strongly associated with aminoglycosides. While, *mph*(A) and *mdt*M genes responsible for macrolide resistance were detected. Regarding the β-lactam resistance, $bla_{\text{TEM-1}}$ gene is associated with resistance to almost penicillins and first generation of cephalosporins (Livermore, 1995). The bla_{AmpC} gene encoding for AmpC β-lactamases confers resistance to a broad spectrum of β-lactams including penicillins, cephalosporins and β-lactamase inhibitor/β-lactam combinations (Jacoby, 2009). Moreover, the presence of $bla_{\text{CTX-M-27}}$ gene underlines the board-spectrum resistance ability towards cephalosporins, carbapenems and penam (Alcock et al., 2020). Like most of other ESBLs and CTX-M family, the CTX-M-27 enzyme is globally distributed (Bonnet *et al.*, 2003). Notably, $bla_{\text{CTX-M-27}}$ becomes the most prominent CTX-M gene in Vietnam (Biedenbach *et al.*, 2014).

Table 2. Antimicrobial resistance genes found in the genome of *E. coli* strain XP817.

Resistance mechanism	AMR gene family	Predicted gene	Resistant to
Antibiotic efflux	Resistance-nodulation-cell division (RND)	*mdt*A, *mdt*B, *mdt*C	Aminocoumarin antibiotic
		*cpx*A, *bae*R, *bae*S, *acr*D	Aminoglycoside
		*mdt*E, *mdt*F, *gad*W, *gad*X, *crp*	Macrolide; fluoroquinolone; penam
		hns, *evg*A, *evg*S	Macrolide; fluoroquinolone; cephalosporin; cephamycin; penam; tetracycline
		*acr*F	Fluoroquinolone; cephalosporin; cephamycin; penam
		*acr*A, *acr*B, *acr*S, *acr*R	Fluoroquinolone; cephalosporin; glycylcycline; penam; tetracycline; rifamycin; phenicol; triclosan
		*mar*R, *sox*R	Fluoroquinolone; cephalosporin; glycylcycline; penam; tetracycline; rifamycin; phenicol; triclosan
		*mar*A, *sox*S	Fluoroquinolone; monobactam; carbapenem; cephalosporin;

			glycylcycline; cephamycin; penam; tetracycline; rifamycin; phenicol; triclosan; penem
		*tol*C	Macrolide; fluoroquinolone; aminoglycoside; carbapenem; cephalosporin; glycylcycline; cephamycin; penam; tetracycline; peptide antibiotic; aminocoumarin; rifamycin; phenicol; triclosan; penem
	Major facilitator superfamily (MFS)	*emr*A, *emr*B, *emr*R, *mdt*H	Nalidixic acid
		*mdt*G	Fosfomycin
		*kpn*E , *kpn*F	Macrolide; aminoglycoside; cephalosporin; tetracycline; peptide; rifamycin
		*mdt*N, *mdt*O, *mdt*P	Nucleoside antibiotic; acridine dye
		tet(A), ***tet*R**, *emr*Y, *emr*K	Tetracycline
		***mdt*M**, *mdf*A	Phenicol
		*evg*A, *evg*S, *hns*, *sox*R, *tol*C	Same as above
	Small multidrug resistance (SMR)		
	ATP-binding cassette (ABC)	*msb*A	Nitroimidazole
		*Yoj*I	Peptide
		*tol*C, *sox*R, *sox*S	Same as above
	KdpDE	*kdp*E	Aminoglycoside
Antibiotic inactivation		***aac*C2**	Aminoglycoside
		$\boldsymbol{ble_{MBL}}$	Glycopeptide
		$\boldsymbol{bla_{CTX\text{-}M\text{-}27}}$, $\boldsymbol{bla_{AmpC}}$, *amp*H	Cephalosporin; penam
		***mph*A**	Macrolide
		$\boldsymbol{bla_{NDM\text{-}1}}$	Carbapenem; cephalosporin; cephamycin; penam
		$\boldsymbol{bla_{TEM\text{-}1}}$	Monobactam; cephalosporin; penam; penem

Antibiotic target alteration	***rmt*B**	Aminoglycoside
	EF-Tu R234F	Elfamycin
	*glp*T E448K	Fosfomycin
	***gyr*A D87N, S83L**; ***par*C S80I**	Fluoroquinolone
	*bac*A, *ept*A, *pmr*F	Peptide

Note: All genes and mutations were documented in the CARD-RGI database. Genes in bold are strongly associated with resistance to important antibiotic groups.

Several genes encoded narrow antibiotic efflux pumps, e.g. *mdt*M (chloramphenicol), *tetA* and *tetR* (tetracycline) were found in *E. coli* XP817. Some constituted versatile exporters as known as MDR efflux pumps, exemplified by tripartite complexes like acrAB-TolC, acrEF-TolC or mdtEF-TolC. The database also reported transcriptional regulators of these pumps as ARGs (e.g. *evg*A, *evg*S, *hns*, *mar*A, *sox*S and *sox*R), which participated significantly in MDR. Furthermore, antibiotic target alteration also contributed greatly to *E. coli* XP817 survival. Mutations in the quinolone-resistance determining region (QRDR) of *gyr*A or *par*C gene are strongly associated with resistance to fluoroquinolones in Gram-negative bacteria. In this study, variants *gyr*A D87N, S83L and *par*C S80I found in strain XP817 are linked to fluoroquinolones resistance, which had been demonstrated previously (Huseby et al., 2017). Similarly, mutation *glp*T E448K found in this strain reduces the import of fosfomycin, consequently led to the resistance (Takahata et al., 2010). Lastly, the detection of $bla_{\text{NDM-1}}$gene in the genome of strain XP817 demonstrates its resistance ability against three carbapenems. Globally, more than 30% of carbapenem-resistant *Enterobacteriaceae* carried the $bla_{\text{NDM-1}}$ (Cui, Zhang, & Du, 2019). Furthermore, we also found ble_{MBL} gene downstream the $bla_{\text{NDM-1}}$ gene causing decreased susceptibility to bleomycin and bleomycin-like molecules. This cluster is observed frequently warning the pressure of anticancer usage can promote the NDM-1 selection and incidentally encourage carbapenemase dissemination (Dortet, Nordmann, & Poirel, 2012).

Plasmid-borne ARGs conferred various antibiotic resistances

Plasmid is the most important mobile genetic element (MGE) that facilitates the transmission of ARGs within and between bacterial species. Thus, establishing a plasmidome would shed more insights into the ARG dissemination. In this study, we detected 05 plasmids in the *E. coli* XP817 which have originated from different species of the *Enterobacteriaceae* family including pNDM1_045001 from *Enterobacter hormaechei* subsp. *Xiangfangensis* WCHEX045001 carrying $bla_{\text{NDM-1}}$ and ble_{MBL}, pMH13-009N_1 from *Proteus mirabilis* MH13-009N carrying *tet(*A*)* and *tet*R, p23141-2 from *Raoultella ornithinolytica* 23141 carrying $bla_{\text{CTX-M-27}}$, pTB211 from *Escherichia coli* ECCHD184 carrying *mph(*A*)*, and pMS14393B *Klebsiella pneumonia* MS14393 *aac*C (Figure 2). These findings underline that *E. coli* XP817 acquired these resistance plasmids via HGTs to become the XDR

phenotype. Hirabayashi et al. 2020 analyzed twenty-five CRE isolates from medical institution in Hanoi and detected *P. mirabilis* strain MH13-009N carrying the plasmid pMH13-009N_1 (Hirabayashi et al., 2020) which is also detected in the plasmidome of the strain XP817. Moreover, *E. hormaechei* pNDM1_045001 carrying *aacC2e* is indicated as a CRE as well and properly transfers ARGs with carbapenem-resistance *K. pneumoniae* strains in Vietnam. In addition, the ST617 is also associated with novel plasmid-borne NDM variants is detected frequently in patients after admitting to the ICU in the hospital system in China (Zong et al., 2018). A recent study showed a high prevalence of colistin-resistant *E. coli* carrying *mcr*-1 gene on chromosome, in which the ST10 was also detected (Yamaguchi et al., 2020). These findings suggest that the wide dissemination of MDR and XDR *E. coli* strains particularly the ST10 clones in the community is the urgent public health issue of Vietnam. Therefore, studying molecular epidemiology will provide comprehensive information of the pathogenic background of *E. coli* strains regarding host identity, growing niche, geographic distribution as well as virulence, resistome and plasmidome profiles.

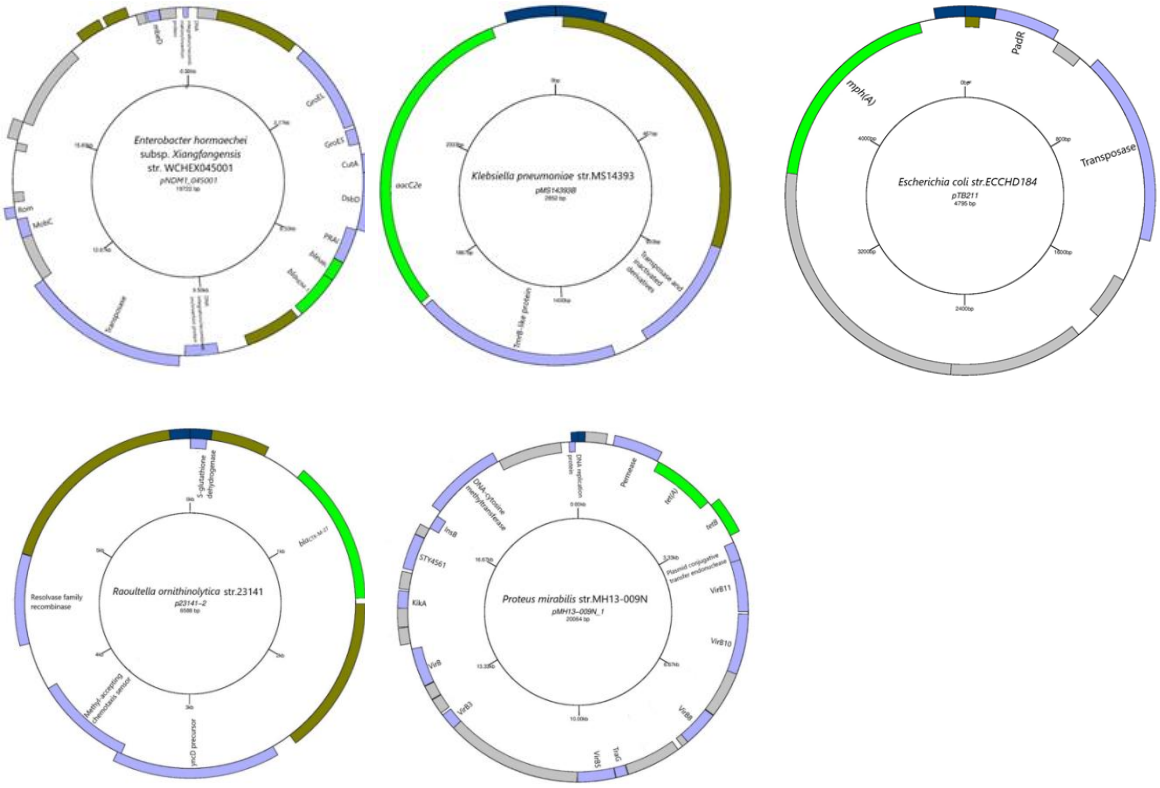

Figure 2. Illustration of five plasmids carrying ARGs in *E. coli* XP817. Color boxes outside the ring present annotated genes on positive strand, and vice versa. Dark green: mobile element protein; Dark blue: repeat region; Gray: hypothetical protein; Light green: annotated ARGs; Light blue: other gene function.

CONCLUSION

The concordance between XDR phenotype and genotype of *E. coli* XP817, suggesting that the WGS would be a powerful tool for the antibiotic resistance surveillance in clinical *E. coli* strains. The finding in this study highlights that *E. coli* is the important reservoir for receiving and disseminating MGEs like plasmids carrying ARGs among *Enterobacteriaceae* family. Finally, the presence of plasmid carrying $bla_{\text{NDM-1}}$ associated with the carbapenem resistance demonstrating that the wide dissemination of carbapenem-resistant *E. coli* strains in Vietnam.

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