Characterization of the plasmid of incompatibility groups IncFII_{pKF727591} and Inc_{pKPHS1} from Enterobacteriaceae species

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Background: Multiple incompatibility (Inc) groups of plasmids have been identified in Enterobacteriaceae species, but there are still quite a few sequenced plasmids that could not be assigned to any known Inc groups.

Methods: One IncFII_{pKF727591}β plasmid p205880-qnrS and two Inc_{pKPHS1} plasmids p11219-CTXM and p205880-NR1 were fully sequenced in this work. Detailed genomic comparison was applied to all available sequenced plasmids of IncFII_{pKF727591} or Inc_{pKPHS1} group.

Results: p205880-qnrS carried a novel transposon Tn6396, which was an ISKpn19-composite transposon and represented a prototype transposable element carrying a minimum core qnrS1 module. p11219-CTXM harbored a novel transposon Tn6559, which was generated from integration of a truncated IS903D-blaCTX-M-14-1SEcp1 unit into the Tn3-family cryptic unit transposon Tn1722. Two Inc groups, IncFII_{pKF727591} and Inc_{pKPHS1}, of plasmids from Enterobacteriaceae species were proposed, and IncFII_{pKF727591} was further grouped into two subgroups IncFII_{pKF727591}α and IncFII_{pKF727591}β. Each of the 11 IncFII_{pKF727591} plasmids carried multiple accessory modules including at least one resistance module, and the relatively small IncFII_{pKF727591} backbones could acquire a wealth of foreign genetic contents. The modular structures of plasmid backbones were conserved within each of IncFII_{pKF727591}α and IncFII_{pKF727591}β subgroups but dramatically different, although with similar gene organizations, between these two subgroups. The Inc_{pKPHS1} backbones were conserved with respect to modular structures, and only four of the 14 Inc_{pKPHS1} plasmids carried accessory modules, two of which contained resistance genes.

Conclusion: A genomic comparison of sequenced Inc_{pKPHS1} or IncFII_{pKF727591} plasmids provides insights into modular differences and genetic diversification of these plasmids, some of which carries antimicrobial resistance genes.

Keywords: plasmids, IncFII_{pKF727591}, Inc_{pKPHS1}, Tn6396, Tn6559

Introduction

Plasmid is a small DNA molecule within a bacterial cell and capable of replicating independently from the host’s chromosomal DNA. Plasmids are mobile genetic elements that commonly carry antimicrobial resistance genes and other genetic factors such as virulence genes. Plasmid-mediated transmission of antimicrobial resistance genes among Enterobacteriaceae and other bacteria imposes a major public health concern.

The original replicon-based scheme to classify plasmids into different incompatibility (Inc) groups was developed in 1970s, which is based on the experimental observations that plasmids with similar replication machinery are often unable to
stably co-exist within the same host cell and thus the plasmid shows incompatibility with the same Inc group plasmid. Nowadays, Inc classification is always based on replication initiation protein (Rep) sequences, and it is not necessarily confirmed by conventional conjugation-based incompatibility experiments. At least 27 Inc groups have been identified in Enterobacteriaceae species, but there are quite a few sequenced plasmids that could not be assigned to any known Inc groups.

This study presented three sequenced plasmids (p205880-qnrS carrying a novel ISKpn19-composite transposon Tn6396, p11219-CTXM harboring a novel Tn722-derivated unit transposon Tn6559, and p205880-NR1 containing no resistance genes) and proposed two novel Inc groups (IncFII<sub>pKF727591</sub> and Inc<sub>pKF727591</sub>). p205880-qnrS belonged to Inc<sub>pKF727591</sub>, while p11219-CTXM and p205880-NR1 could be assigned to IncFII<sub>pKF727591</sub>. Further detailed genomic comparison of all sequenced plasmids of Inc<sub>pKF727591</sub> or IncFII<sub>pKF727591</sub> indicated considerable modular differences and genetic diversification of each group of plasmids.

**Materials and methods**

**Bacterial strains and genome sequencing**

*Klebsiella pneumoniae* 205880 and 11219 were recovered from the sputum specimens of two different patients with pneumonia in two different Chinese hospitals in 2012 and 2013, respectively. For each strain, genomic DNA isolation, genome sequencing, and sequence assembly and annotation were carried out as described previously. An unrooted neighbor-joining tree was generated from the aligned *repA* sequences of indicative plasmids. Plasmids p205880-qnrS, p11219-CTXM and p205880-NR1 had GenBank accession numbers MF190368, MF133442 and MF144193, respectively.

**Phenotypic assays**

Plasmid conjugal transfer was carried out, as described previously, with *Escherichia coli* EC600 as a recipient and the 205880 or 11219 isolates as a donor, for selecting an *E. coli* transconjugant that carried bla<sub>CTX-M-14</sub> (p11219-CTXM) or qnrS1 (p205880-qnrS), respectively. Electroporation of plasmid p11219-CTXM from the 11219 isolate into *E. coli* TOP10 was performed, as described previously, to obtain an *E. coli* electroporant carrying bla<sub>CTX-M-14</sub> (p11219-CTXM). Double-disk synergy test was performed to detect the activity of extended-spectrum β-lactamase (ESBL) in indicative bacterial strains.

**Results and discussion**

**Diversification of IncFII<sub>pKF727591</sub> plasmids**

One new plasmid p205880-qnrS was fully sequenced (Table 1 and could be transferred from the wild-type 205880 isolate into EC600, through conjugation, giving a qnrS-positive transconjugant p205880-qnrS-EC600. As expected, these two strains were resistant to ciprofloxacin and levofloxacin with minimum inhibitory concentration (MIC) values ≥4.

A collection of 11 plasmids including p205880-qnrS (Table S1), which had homologous *repA* (replication initiation) genes and similar backbone gene organizations, were assigned into a novel Inc group designated IncFII<sub>pKF727591</sub> (Inc<sub>reference plasmid</sub>), because all these *repA* proteins had an IncFII super-family domain. The phylogenetic tree (Figure 1) based on *repA* sequences indicated that these 11 plasmids could be divided into two separately clustering subgroups IncFII<sub>pKF727591</sub>α (n=8) and IncFII<sub>pKF727591</sub>β (n=3). As shown by pairwise comparison of *repA* nucleotide sequences, plasmids within each subgroup showed 100% identity, while those from different subgroups displayed ≥79% identity (Table S2A). Predicted RepA-binding iterons were located from 245 bp to 365 bp downstream of *repA* for IncFII<sub>pKF727591</sub>α plasmids, but upstream from 366 bp to 460 bp for IncFII<sub>pKF727591</sub>β plasmids, and three copy numbers of iteron were found for all IncFII<sub>pKF727591</sub> plasmids (Table S1). Plasmids within each subgroup shared a conserved iteron motif, but those from different subgroups had dramatically different iteron motifs (Figure 1).

pKF727591 (the first sequenced IncFII<sub>pKF727591</sub> plasmid) and pKp_Goe_414-4 (the first sequenced IncFII<sub>pKF727591</sub>β plasmid) were identified as the references for IncFII<sub>pKF727591</sub>α and IncFII<sub>pKF727591</sub>β, respectively. p205880-qnrS belonged to IncFII<sub>pKF727591</sub>β.

The modular structure (Table 1 and Figure S1) of each plasmid could be divided into one or more accessory modules (defined as acquired DNA regions associated or bordered with mobile elements) and the remaining IncFII<sub>pKF727591</sub> backbone regions (responsible for plasmid replication, maintenance and conjugal transfer). The eight IncFII<sub>pKF727591</sub>α plasmids shared ≥88% of their backbone sequences with ≥99% nucleotide identity, and the three IncFII<sub>pKF727591</sub>β plasmids showed ≥99% nucleotide
| Inc group | Plasmid | Accession number | Total length (bp) | Total number of open reading frames | Mean G+C content, % | Length of the backbone (bp) | Accessory module(s) | Reference |
|-----------|---------|------------------|------------------|-------------------------------------|---------------------|-----------------------------|--------------------|-----------|
| IncFII    | pKF727591 | KF727591         | 94,790           | 112                                 | 53.1                | 51,559                      | NDM-1 region<sup>‡</sup>, and ΔISEcl6 | NA        |
|           | pKpn235-BG | KT852336         | 76,360           | 90                                  | 53.9                | 51,749                      | NDM-1 region<sup>‡</sup>, and ΔISEcl6 | NA        |
|           | pKpn240-BG | KT852335         | 76,980           | 92                                  | 53.8                | 51,594                      | NDM-1 region<sup>‡</sup>, and ΔISEcl6 | NA        |
|           | pB-3002cz  | KJ958926         | 97,650           | 122                                 | 53.2                | 51,558                      | NDM-1 region<sup>‡</sup>, and ΔISEcl6 | NA        |
|           | pEhA      | KR822246         | 96,120           | 105                                 | 53.1                | 51,559                      | NDM-1 region<sup>‡</sup>, and ΔISEcl6 | NA        |
|           | pCP020050  | CP020050         | 113,430          | 129                                 | 52.5                | 46,102                      | ΔIS<sup>†</sup>, and ΔISEcl6       | NA        |
|           | pLN824135  | LN824135         | 118,320          | 142                                 | 52.5                | 46,102                      | ΔIS<sup>†</sup>, and ΔISEcl6       | NA        |
|           | pCAV1217-71 | CP018674        | 70,610           | 95                                  | 52.4                | 46,100                      | ΔIS<sup>†</sup>, and ΔISEcl6       | NA        |
| IncFII    | pKp_Goe_414-4<sup>‡</sup> | CP018341 | 81,641           | 88                                  | 53.9                | 54,186                      | MDR region<sup>‡</sup>, and ΔIS<sup>†</sup> | NA        |
|           | p205880-qnrS<sup>§</sup> | CP019038 | 65,110           | 75                                  | 52.9                | 54,649                      | IS<sup>§</sup>26–ΔTn<sup>§</sup>1696, and Tn6396<sup>‡</sup> | NA        |
|           | p205880-2  | MF133496         | 79,370           | 83                                  | 54.1                | 54,726                      | MDR region<sup>‡</sup>            | This study |
| IncPHSI   | pKPHSI | CP003223         | 122,800          | 131                                 | 49.5                | 113,828                     | Tn6558 region<sup>‡</sup>          | 14        |
|           | pRJA166c   | CP019050         | 111,080          | 117                                 | 49                  | 111,080                     | None                           | NA        |
|           | pPMK1-B    | CP008931         | 111,690          | 117                                 | 49.2                | 111,690                     | None                           | NA        |
|           | pSgl-1     | CP012427         | 126,470          | 134                                 | 49.2                | 126,470                     | None                           | NA        |
|           | p11219-CTXM | MF133442       | 122,080          | 128                                 | 50                  | 110,993                     | Tn6559 region<sup>‡</sup>, and ISKpn24 | This study |
|           | pCP020063  | CP020063         | 109,020          | 119                                 | 49.2                | 107,422                     | ISKpn28                        | NA        |
|           | pCP015755  | CP015755         | 109,350          | 125                                 | 49.3                | 109,350                     | None                           | NA        |
|           | pCP016161  | CP016161         | 109,350          | 123                                 | 49.3                | 109,350                     | None                           | NA        |
|           | pUCLAOXA232-4 | CP012565      | 112,060          | 117                                 | 49                  | 112,060                     | None                           | NA        |
|           | pUCLAOXA232-4.X | CP012570     | 111,240          | 115                                 | 49                  | 111,240                     | None                           | NA        |
|           | pUCCLAUXA1400954 | CP016924    | 111,540          | 116                                 | 49.3                | 111,540                     | None                           | NA        |
|           | pKPN-04f   | CP014756         | 121,030          | 118                                 | 49.3                | 109,640                     | IS<sup>§</sup>3000, and ISKpn25       | NA        |
|           | p205880-NR1 | MF144193      | 108,040          | 117                                 | 49.3                | 108,040                     | None                           | This study |
|           | pKP301b    | KJ354306         | 110,253          | 111                                 | 48.3                | 110,653                     | None                           | NA        |

Notes: p205880-qnrS, p11219-CTXM and p205880-NR1 were sequenced in this study, while all the other plasmids were derived from GenBank (last accessed May 28, 2017). ‡Reference plasmids. †Carrying resistance gene.

Abbreviation: NA, not applicable.
identity over ≥98% of their backbone sequences; by contrast, the backbones of IncFII<sub>pKF727591</sub>α and IncFII<sub>pKF727591</sub>β had ≤92% nucleotide identity across ≤70% of their backbone sequences (Table S2B). The modular structures of plasmid backbones were conserved within each of IncFII<sub>pKF727591</sub>α and IncFII<sub>pKF727591</sub>β subgroups but dramatically different between two subgroups.

Integration of accessory modules at various sites of IncFII<sub>pKF727591</sub> backbones led to the interruption of relevant backbone genes (e.g., umuC), the disruption of the maintenance or conjugal transfer regions, or the deletion of surrounding backbone regions (e.g., 5.3-kb deletion containing mtsM) (Figure 2). The IncFII<sub>pKF727591</sub> replicons and the conjugal transfer regions (encoding an F-type type IV secretion system) were found in all 11 plasmids and thus represented the core IncFII<sub>pKF727591</sub> backbone. An 11-kb maintenance region carrying parAB (partition) and ccdBA (toxin-antitoxin) was found in all IncFII<sub>pKF727591</sub>α plasmids, while another distinct 15-kb maintenance region containing stbAB (mediator of plasmid stability) and resD (resolvase) in all IncFII<sub>pKF727591</sub>β plasmids.

All the three IncFII<sub>pKF727591</sub>β plasmids contained a single IncFII<sub>pKF727591</sub>β replicon, which six of the eight IncFII<sub>pKF727591</sub>α plasmids contained a second Inc<sub>pA1763-KPC</sub> replicon beside the master IncFII<sub>pKF727591</sub>α replicon (Figure 2). Notably, the Inc<sub>pA1763-KPC</sub> replicon was located within a 9.7- or 8.1-kb backbone region [carrying maintenance
genes such as parA and resA; as observed in pA1763-KPC (GenBank accession number MH909340), which was a part of relevant accessory modules (see below; Figure 3). Two coexistent replicons IncFIIpKF72759|α and IncpA1763-KPC, together with their supporting maintenance genes, will promote relevant plasmids to overcome incompatibility barrier with incoming plasmids. All the above replicons belonged to the iteron-regulated replicon, for which Rep monomers specifically bound to iterons. 5

Accessory resistance modules of IncFIIpKF72759|α plasmids

A large accessory module was integrated at a site between the two maintenance genes mtsM and ccdA in each of the eight IncFIIpKF72759|α plasmids (Figure 3). These eight modules had some common regions but showed considerable modular differences across the whole modules, indicating their sole evolutionary origin followed by parallel mosaic diversification. The 9.7- or 8.1-kb backbone region from IncpA1763-KPC (see above) was found in six IncFIIpKF72759|α plasmids except for pKpn235-BG and pKpn240-BG. The accessory modules from seven IncFIIpKF72759|α plasmids, except for pCAV1217-71, carried resistance loci (Figure 3 and Table S3): i) a truncated Tn1725 transposon carrying blaN|6 was harbored in pB-3002cz, pKpn235-BG and pKpn240-BG, while another truncated version of Tn125 in pKF727591 and pEh1A; ii) the ars (arsenical resistance) locus was found in pKF727591, pEh1A and pCP020050 and, notably, the first two plasmids showed coexistence of blaN|6 and ars; and iii) qnrB1 and dfrA14-carrying In191 were identified in pLN824135.

A 24.7-kb MDR region, another 26.9-kb MDR region and Tn6396 (Figure 4) were inserted at the same site within the umuC gene of the three IncFIIpKF72759|β plasmids p675920-2, pKp_Goe_414-4 and p205880-qnrS, respectively. The 24.7-kb MDR region, carrying multiple resistance genes (Table S3), was generated from integration of an IS26ΔTn6346-ΔGlsul2-IS26 unit into Tn1721, which was further connected with a truncated IS26-blaLAP-2–qnrS1–IS26 unit. The 26.9-kb MDR region was highly similar to the 24.7-kb MDR region but differed from it mainly by inversion of IS26–ΔTn6346–ΔGlsul2–IS26 and further upstream insertion of an IS26–pdk–catA2–IS26 unit. Tn6396 was a novel ISKpn19-composite transposon, which carried the qnrS1–ΔtnpR region and bracketed by 7-bp direct repeats (DRs: target site duplication signals for transposition) at both ends. Tn6396 represented a prototype transposable element carrying a minimum core qnrS1 module. Different Tn6396
derivatives with distinct terminal truncations were found in various plasmids including pKp_Goe_414-4 and p675920-2 (Figure S2).

Characterization of Inc_pKPHS1 plasmids

Two additional new plasmids p11219-CTXM (carrying bla_CTXM-14) and p205880-NR1 (containing no resistance genes) were fully sequenced (Table S1 and Figure S1). p11219-CTXM could not be transferred from the wild-type 11219 isolate into EC600 through conjugation, but could be transferred into TOP10 through electroporation, generating a bla_CTXM-14-positive electroporant 11219-CTXM-TOP10. These two wild-type and electroporant strains had ESBL activity (data not shown) and were resistant to cefazolin, cefuroxime and ceftazidime with MIC values ≥64.

A total of 14 plasmids including p11219-CTXM and p205880-NR1 (Table S1), each of which carried a single repA gene with >96% nucleotide identity to repA_pKPHS1 (Table S4A) and had a backbone gene organization similar to pKPHS1 (Figure S5), were assigned into a novel Inc group named as Inc_pKPHS1 (Figure S3). These 14 RepA proteins did not any of known domain super-families. Four copy numbers of a conserved iteron motif (Figure S3) were found 48 bp to 218 bp downstream of repA for all Inc_pKPHS1 plasmids (Table S1). All plasmids carried a single iteron-regulated Inc_pKPHS1 replicon. pKPHS1, the first sequenced Inc_pKPHS1 plasmid, was identified as the Inc_pKPHS1 reference. These 14 plasmids had >96% nucleotide identity over >75% coverage of their backbone sequences (Table S4B). Modular differences were found at multiple sites of the maintenance regions (Figure 5). None of conjugal transfer genes was found in all plasmids, which was consistent to the non-conjugative nature of p11219-CTXM. Remarkably, all these plasmids carried ɸpKPHS1 regions resembling SSU5 phage. Only four Inc_pKPHS1 plasmids had accessory modules, including the two resistance modules: Tn6558 from pKPHS1 and Tn6559 from p11219-CTXM (Table 1 and Figure 5).

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Figure 4 Organization of accessory resistance modules from IncFII_pKF72751 β-plasmids and comparison with related regions. Genes are denoted by arrows. Genes, mobile elements and other features are colored based on function classification. Shading denotes regions of homology (>95% nucleotide identity). Numbers in brackets indicate nucleotide positions within corresponding plasmids. The accession numbers of Gla219, Tn6346, Tn1728 and IS26-bla_CTXM-14- qnrS1-IS26 unit11 for reference are CP001918, EU696790, X61367 and HF545433, respectively.
Figure 5  Linear comparison of complete sequences of IncP1 plasmids. Genes are denoted by arrows. Genes, mobile elements and other features are colored based on function classification. Shading regions denote homology of plasmid backbone regions (≥90% nucleotide identity) but not accessory modules.

Figure 6  Organization of Tn6558 and Tn6559 and comparison with related regions. Genes are denoted by arrows. Genes, mobile elements and other features are colored based on function classification. Shading denotes regions of homology (>95% nucleotide identity). Numbers in brackets indicate the nucleotide positions within the corresponding plasmids. The accession numbers of IS{Ecp1–bla}CTX-M-14–IS903D unit are KX646543 and X61367, respectively.
Ethics approval and informed consent
This study needs not to be reviewed or approved by the ethics committee of the hospitals, because the bacterial isolate involved in this study was part of the routine hospital laboratory procedure. The research involving biohazards and all related procedures were approved by the Biosafety Committee of the Beijing Institute of Microbiology and Epidemiology.

Acknowledgment
This work was supported by the National Key R&D Program (2018YFC1200100) of China and the Local Social Science Project (2018QD0031) of Heilongjiang Province.

Author contributions
All authors contributed toward data analysis, drafting and revising the paper, gave final approval of the version to be published and agree to be accountable for all aspects of the work.

Disclosure
The authors report no conflicts of interest in this work.

References
1. Shintani M, Sanchez ZK, Kimbara K. Genomics of microbial plasmids: classification and identification based on replication and transfer systems and host taxonomy. Front Microbiol. 2015;6:242. doi:10.3389/fmicb.2015.00242
2. Zhan Z, Hu L, Jiang X, et al. Plasmid and chromosomal integration of four novel blaIMP-carrying transposons from Pseudomonas aeruginosa, Klebsiella pneumoniae and an Enterobacter sp. J Antimicrob Chemother. 2018;73(11):3005–3015. doi:10.1093/jac/dky288
3. CLSI. Performance Standards for Antimicrobial Susceptibility Testing: Twenty-Fifth Informational Supplement M100-S25. Wayne (PA): Clinical and Laboratory Standards Institute; 2015.
4. Wayne. Performance Standards for Antimicrobial Susceptibility Testing: Twenty-seventh Informational Supplement M100-S27. CLSI; 2017.
5. Pilla G, Tang CM. Going around in circles: virulence plasmids in enteric pathogens. Nat Rev Microbiol. 2018;16(8):484–495. doi:10.1038/s41579-018-0031-2
6. Poirel L, Bonnin RA, Boulanger A, Schrenzel J, Kaase M, Nordmann P. Tn125-related acquisition of blaNDM-like genes in Acinetobacter baumannii. Antimicrob Agents Chemother. 2012;56(2):1087–1089. doi:10.1128/AAC.05620-11
7. Feng J, Yin Z, Zhan Z, et al. Structure genomics of two chimera plasmids p675920-1 and p675920-2 coexisting in a multi-drug resistant Klebsiella pneumoniae isolate. Oncotarget. 2018. doi:10.18632/oncotarget.24235
8. Allmeier H, Crezan B, Greck M, Schmitt R. Complete nucleotide sequence of Tn1721: gene organization and a novel gene product with features of a chemotaxis protein. Gene. 1992;111(1):11–20. doi:10.1016/0378-1119(92)90597-4
9. Kim M, Kim S, Ryu S. Complete genome sequence of bacteriophage SSU5 specific for Salmonella enterica serovar Typhimurium rough strains. J Virol. 2012;86(19):10894. doi:10.1128/JVI.01796-12
10. Poirel L, Dartus MF, Decouwer JW, Nordmann P. ISEcp1B-mediated transposition of blaCTX-M in Escherichia coli. Antimicrob Agents Chemother. 2005;49(1):447–450. doi:10.1128/AAC.49.1.447–450.2005
11. Studentova V, Dobiasova H, Hedlova D, Dolejska M, Papagiannitsis CC, Hrabak J. Complete nucleotide sequences of two NDM-1-encoding plasmids from the same sequence type 11 Klebsiella pneumoniae strain. Antimicrob Agents Chemother. 2015;59(2):1325–1328. doi:10.1128/AAC.04095-14
12. Papagiannitsis CC, Malli E, Florou Z, et al. Emergence of sequence type 11 Klebsiella pneumoniae coproducing NDM-1 and VIM-1 metallo-beta-lactamases in a Greek hospital. Diagn Microbiol Infect Dis. 2017;87(3):295–297. doi:10.1016/j.diagmicrobio.2016.12.008
13. Campos JC, Da Silva MJ, Dos Santos PR, et al. Characterization of Tn3000, a transposon responsible for blaNDM-1 dissemination among enterobacteriaceae in Brazil, Nepal, Morocco, and India. Antimicrob Agents Chemother. 2015;59(12):7387–7395. doi:10.1128/AAC.01458-15
14. Liu P, Li P, Jiang X, et al. Complete genome sequence of Klebsiella pneumoniae subsp. pneumoniae HS1286, a multidrug-resistant strain isolated from human sputum. J Bacteriol. 2012;194(7):1841–1842. doi:10.1128/JB.00043-12
15. Stoesser N, Giess A, Batty EM, et al. Genome sequencing of an extended series of NDM-producing Klebsiella pneumoniae isolates from neonatal infections in a Nepali hospital characterizes the extent of community- versus hospital-associated transmission in an endemic setting. Antimicrob Agents Chemother. 2014;58(12):7347–7357. doi:10.1128/AAC.03900-14
16. Khong WX, Marimuthu K, Teo J, et al. Tracking inter-institutional spread of NDM and identification of a novel NDM-positive plasmid, pSg1-NDM, using next-generation sequencing approaches. J Antimicrob Chemother. 2016;71(11):3081–3089. doi:10.1093/jac/dkw277
17. Antonelli A, D’Andrea MM, Vaggelli G, Docquier JD, Rossolini GM. OXA-372, a novel carbapenem-hydrolysing class D beta-lactamase from a Citrobacter freundii isolated from a hospital wastewater plant. J Antimicrob Chemother. 2015;70(10):2749–2756. doi:10.1093/jac/dkw181
18. Espedido BA, Steen JA, Ziochos H, et al. Whole genome sequence analysis of the first Australian OXA-48-producing outbreak-associated Klebsiella pneumoniae isolates: the resistome and in vivo evolution. PLoS One. 2013;8(3):e59920. doi:10.1371/journal.pone.0059920
19. Nigro SJ, Hall RM. GsLu2, a genomic island carrying the sul2 sulphonamide resistance gene and the small mobile element CR2 found in the Enterobacter cloacae subspecies cloacae type strain ATCC 13047 from 1890, Shigella flexneri ATCC 700930 from 1954 and Acinetobacter baumannii ATCC 17978 from 1951. J Antimicrob Chemother. 2011;66(9):2175–2176. doi:10.1093/jac/dkr230
20. Ng SP, Davis B, Palombo EA, Bhave M. A Tn5051-like mer-containing transposon identified in a heavy metal tolerant strain Achromobacter sp. AO22. BMC Res Notes. 2009;2:38. doi:10.1186/1756-0500-2-38
21. Le V, Nhu NT, Cerdeno-Tarraga A, et al. Genetic characterization of three qnrS1-harbouring multidrug-resistance plasmids and qnrS1-containing transposons circulating in Ho Chi Minh City, Vietnam. J Med Microbiol. 2015;64(8):869–878. doi:10.1099/jmm.0.000100
22. Wang L, Liu L, Liu D, et al. The first report of a fully sequenced resistance plasmid from Shigella boydii. Front Microbiol. 2016;7:1579. doi:10.3389/fmicb.2016.01579
