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GENOME SEQUENCES

Draft Genome Sequence of an Extended-Spectrum β-Lactamase-Producing *Klebsiella oxytoca* Strain Bearing *mcr-9* from Qatar

Clement K. M. Tsui,[a,b,c] Sathyavathi Sundararaju,[a] Hassan Al Mana,[a,d] Mohammad Rubayet Hasan,[a,b] Patrick Tang,[a,b] Andres Perez-Lopez[a,b]

[a]Department of Pathology, Sidra Medicine, Doha, Qatar
[b]Department of Pathology and Laboratory Medicine, Weill Cornell Medicine-Qatar, Doha, Qatar
[c]Division of Infectious Diseases, Faculty of Medicine, University of British Columbia, Vancouver, BC, Canada
[d]Biomedical Research Center, Qatar University, Doha, Qatar

ABSTRACT *Klebsiella oxytoca* is an opportunistic human pathogen causing nosocomial infection. We report the draft genome of an extended-spectrum β-lactamase-producing *K. oxytoca* isolate harboring an *mcr-9* gene, a recently discovered colistin resistance analog, from Qatar. The genome statistics, along with the sequence type and resistance mechanisms, are predicted for the assembled genome.

Colistin in combination with other agents is used for the treatment of severe infections caused by carbapenem-resistant Gram-negative bacteria. The emergence of plasmid-mediated resistance to colistin due to lipid A-modifying enzymes encoded by 10 different *mcr* genes in *Enterobacterales* has endangered the last-resort treatment (1–3). One of the genes, *mcr-9*, has been recently described in *Escherichia coli*, *Klebsiella pneumoniae*, *Enterobacter* spp., and *Salmonella enterica* serotype Typhimurium (4–9) from animals and humans. The increasing reports of novel *mcr* genes have become a great concern in many countries. Therefore, it is crucial to understand the molecular mechanisms of colistin resistance in Qatar.

Klebsiella oxytoca is an opportunistic pathogen associated with nosocomial infections (10, 11). It becomes a public health concern because of resistance to antimicrobials due to the presence of bla_{OXY}, $bla_{CTX\text{-}M}$, and bla_{SHV} genes (11). Here, we report the genome of *K. oxytoca* strain 18099069b harboring *mcr-9*, which was isolated from a rectal swab obtained from a child during admission to the intensive care unit to screen for extended-spectrum β-lactamase (ESBL)- and carbapenemase-producing *Enterobacteriaceae*. Swabs are routinely inoculated onto CHROMagar ESBL and CHROMagar mSuperCARBA (CHROMagar, France). Plates are incubated under aerobic conditions at 35 ± 2°C for 18 to 24 h, minimizing exposure to light. Suspicious colonies growing on the chromogenic agar plates after overnight incubation are identified using the matrix-assisted laser desorption ionization–time of flight (MALDI-TOF) Biotyper system (Bruker, Bremen, Germany). In this case, MALDI-TOF mass spectrometric identification was performed on a single colony picked from the ESBL chromogenic plate onto the MALDI target plate. Antimicrobial susceptibility testing was performed using the Phoenix system (Becton, Dickinson, USA). The MIC for colistin was determined by broth microdilution (ComASP Colistin; Liofilchem, Italy). MICs were interpreted according to the CLSI breakpoints (12). Ethics approval for the study was obtained from the institutional review board of Sidra Medicine.

DNA was extracted using the NucliSENS easyMag platform (bioMérieux, France). DNA libraries were constructed with a Nextera XT kit (Illumina, CA) and sequenced on the Illumina MiSeq system with 2 × 300 cycles. The sequence data were trimmed using

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Address correspondence to Clement K. M. Tsui, ktsui@sidra.org.

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TABLE 1 Summary of genome statistics and genetic mechanism of antibiotic resistance

Parameter	Observation
Isolate	18099069b
No. of reads	1,526,962
Genome size (bp)	6,190,059
N_{50} (bp)	210,467
No. of contigs	95
Avg coverage (×)	135
GC content (%)	54.8
No. of coding sequences	5,792
ST	58
Plasmid types	IncHI2, IncHI2A, IncFIB(K)
AMR genes	bla_{OXY2-2}, bla_{SHV-12}, bla_{TEM-1B}, *aac(6′)-Ilc*, *aph(3′)-Ia*, *aac(3)-IIb*, *ere*(A), *oqxAB*, *fosA*, *sul1*, *sul2*, *catA2*
Antibiotic resistance phenotypes[a]	AMP, CAZ, PIP, GEN, SXT

[a] AMP, ampicillin; CAZ, ceftazidime; PIP, piperacillin; GEN, gentamicin; SXT, trimethoprim-sulfamethoxazole.

Trim Galore v0.6.1 (http://www.bioinformatics.babraham.ac.uk/projects/trim_galore), assembled using SPAdes v3.9.0 (13), and assessed using QUAST v5.0.2 (14). Contaminant data were analyzed by Kraken v2 (15). The sequence type (ST), plasmids, and antimicrobial resistance (AMR) genes were predicted using the mlst (https://github.com/tseemann/mlst), PlasmidFinder v2.1 (16), and ResFinder v3.2 (17) databases in abricate v0.9.8 (https://github.com/tseemann/abricate), based on ≥60% coverage and 90% sequence identity. The repeat sequences of strains were identified using ISfinder v2 (18). The assembled genome was annotated by the NCBI Prokaryotic Genome Annotation Pipeline v4.11 (19). Default parameters were used for all software unless otherwise specified.

The genome statistics for 18099069b are summarized in Table 1. This isolate contained bla_{TEM1B}, bla_{SHV12}, and bla_{OXY22}, encoding different class A β-lactamases, which confer resistance to not only narrow-spectrum β-lactams but also broad-spectrum cephalosporins (Table 1). This strain was susceptible to colistin with a MIC of 2 μg/ml, consistent with the report that resistance to colistin (MIC, ≥4 μg/ml) in the presence of *mcr-9* occurs only when the regulatory genes *qseC* and *qseB* are expressed (5). A BLAST search of the contig (2,659 bp) containing *mcr-9* indicated 100% identity to plasmids of multiple bacteria; two of them (GenBank accession numbers CP043767.1 and CP044215.1) were annotated as replicon type IncHI2, which is well known for its association with bla_{SHV-12} and *mcr-9* (6). Insertional elements IS*903* and IS*15* were also detected in the flanking regions of *mcr-9*.

Data availability. This whole-genome shotgun project has been deposited in DDBJ/ENA/GenBank under the accession number JAAWXG000000000, as well as accession numbers PRJNA577539 and SRR11485633. The version described in this paper is version JAAWXG010000000.

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