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GENOME SEQUENCES

# Complete Genome Sequence of a Carbapenem-Resistant *Escherichia coli* Isolate with $bla_{NDM-5}$ from a Dog in the United States

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**ABSTRACT** The carbapenem resistance gene $bla_{NDM-5}$ was identified in an *Escherichia coli* strain isolated from a dog. We report here the complete genome sequence of this *E. coli* strain; the $bla_{NDM-5}$ gene was present on a large IncFII multidrug-resistant plasmid. This is the first $bla_{NDM-5}$-carrying *E. coli* strain from an animal in the United States.

Carbapenems are a class of beta-lactam antibiotics that are active against many aerobic and anaerobic Gram-positive and Gram-negative organisms. They are critically important antibiotics used to treat serious bacterial infections (1). Carbapenem resistance is rare and typically results from expression of a carbapenemase that hydrolyzes penicillins, cephalosporins, monobactams, and carbapenems. Known genes include $bla_{KPC}$, $bla_{OXA}$, $bla_{VIM}$, and $bla_{NDM}$, among others, and have been found in various Gram-negative organisms (2–4).

In July 2018, *Escherichia coli* strain 24213-18 was isolated from an endotracheal wash specimen obtained from a dog in a veterinary hospital in Philadelphia, Pennsylvania. The isolate was phenotypically resistant to imipenem (MIC, 4 $\mu$g/ml), and short-read sequencing identified the carbapenem resistance gene $bla_{NDM-5}$. As a result, the isolate was sequenced by long-read technology.

The isolate was grown on blood agar at 35°C for 24 h. Genomic DNA was extracted with the DNeasy blood and tissue kit (Qiagen, Germantown, MD), quantified on the Qubit fluorometer with the Qubit double-stranded DNA (dsDNA) broad-range (BR) assay kit, and fragmented to 10 kb by using a G-tube. The quality and size of DNA were measured by FEMTO Pulse. The DNA library, ligated with a symmetrically paired barcode, was prepared following the 10-kb template preparation protocol with SMRTbell template prep kit v 1.0. Whole-genome sequencing (WGS) was performed on a Pacific Biosciences Sequel sequencer with Sequel sequencing kit v 3.0. Sequences were collected after 120 min of preextension for 600 min of movie length.

There were 65,426 polymerase reads with a 41,595-bp mean read length and 620,057 subreads with an $N_{50}$ value of 6,330 bp. The reads were filtered with high-quality read filtering at the primary analysis step, to ensure that only single reads longer than 50 bp per zero-mode waveguide were used, and were *de novo* assembled by the PacBio Hierarchical Genome Assembly Process (HGAP) v 4.0 program. Each assembled contig had a mean confidence quality value above 90 (one error in 1 Gb). The contigs were further circularized with Circlator (5) with the default parameters. The sequences were annotated on the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) v 4.8 (6).

The total genome size was 5,135,313 bp with a GC content of 50.8%. Assembly resulted in a single circular chromosome of 4,857,938 bp, with 93× coverage, and two plasmids. WGS-based multilocus sequence typing (MLST) (7) determined that this *E. coli*

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strain was sequence type 167 (ST167), the same sequence type as that of dog isolates with $bla_{NDM-5}$ from Finland (8). There was a circular IncFII plasmid of 139,547 bp with 93× coverage that contained the $bla_{NDM-5}$ gene and the additional resistance genes *tet*(A), *aac(6′)-Ib-cr*, *aadA5*, *aadA2*, $bla_{OXA-1}$, $bla_{CTX-M-15}$, *catB3*, *dfrA7*, *dfrA12*, *sul1* (two copies), and *mph*(A). These genes confer resistance to various drug classes, and confirmed phenotypic testing on a Vitek 2 system (bioMérieux) using the GN65 panel showed resistance to tetracycline, aminoglycosides, beta-lactams, chloramphenicol, fluoroquinolones, and trimethoprim-sulfamethoxazole. This plasmid is highly related to plasmid p28078-NDM found in *E. coli* from China (GenBank accession number MF156713). The additional contig was not circularized, and it was an IncI plasmid of 137,828 bp with 91× coverage and resistance genes $bla_{TEM-1D}$, *floR*, *ant(3″)-Ia*, and *aac(3)-IId*.

**Data availability.** The assembled genome sequence for this isolate was submitted to GenBank under accession numbers CP041392, CP041393, and CP041394. Raw sequence reads were deposited into the SRA under accession number SRR9668343.

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