Draft Genome Sequence of *Raoultella planticola*, Isolated from River Water

Narayanan Jothikumar, a Amy Kahler, a Nancy Stockbine, a Lori Gladney, a,b Vincent R. Hill a

National Center for Emerging and Zoonotic Infectious Diseases, Centers for Disease Control and Prevention, Atlanta, Georgia, USA a; IHRC, Inc., Atlanta, Georgia, USA a,b

We isolated *Raoultella planticola* from a river water sample, which was phenotypically indistinguishable from *Escherichia coli* on MI agar. The genome sequence of *R. planticola* was determined to gain information about its metabolic functions contributing to its false positive appearance of *E. coli* on MI agar. We report the first whole genome sequence of *Raoultella planticola*.

Received 8 September 2014 Accepted 10 September 2014 Published 16 October 2014

Citation Jothikumar N, Kahler A, Stockbine N, Gladney L, Hill VR. 2014. Draft genome sequence of *Raoultella planticola*, isolated from river water. Genome Announc. 2(5): e01061-14. doi:10.1128/genomeA.01061-14.

Copyright © 2014 Jothikumar et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 3.0 Unported license.

Address correspondence to Narayanan Jothikumar, jin2@cdc.gov.

U.S. drinking water regulations require that water samples be examined for fecal contamination using U.S. EPA-approved methods. Cultivation of membrane filtered water samples on MI agar has been used as a reliable method for detecting such contamination because of its ability to simultaneously detect and quantify both total coliforms and *Escherichia coli* specifically. MI agar is a selective and differential medium, containing cefsulodin to inhibit Gram-positive bacteria and noncoliform Gram-negative bacteria and indoxyl-β-D-glucuronide and 4-methylumbelliferyl-β-D-galactopyranoside for the presumptive identification of *E. coli* and other coliform bacteria, respectively (1). Although most coliform bacteria other than *E. coli* do not express β-glucuronidase activity, some strains of *Klebsiella* have been reported to express this enzyme and appear indistinguishable from *E. coli* on MI agar (2, 3). It is important to identify false-positive bacteria on MI agar to improve water quality monitoring techniques and risk assessments for the fecal contamination of water. A suspect blue colony of *E. coli* was isolated from a river water sample in 2012 on MI agar and was identified as *R. planticola* by *rpoB* gene sequencing (4). To understand the metabolic function of *R. planticola* strain CHB, whole genome sequencing was performed. The isolate was cultured in 10 mL Luria-Bertani broth overnight at 37°C. The overnight culture of 1 to 5×10^8 CFU/mL was pelleted and resuspended in 350 μL PBS. The genomic DNA was extracted by mixing with an equal amount of 2×UNEX nucleic acid extraction buffer (Microbio- logics, MN) as per manufacturer’s instructions followed by a final purification step using a polyvinylpyrrolidone (PVPP) spin column (Spin-IV-HRC columns, Zymo Research Corporation, Orange, CA).

The whole genome was sequenced using the Illumina MiSeq for paired-end 300×300 library preparation (MR DNA, Shal- lowater, TX). The genome sequence was assembled using DNASTar SeqMan NGen resulting in 18 contigs and a total length of 5.8 Mb. The genome had coverage of 422× (N_{50} of 561 Kb) and a G+C content of 55.4%. Annotation was performed using the RAST (Rapid Annotation using Subsystems Technology) server (5). The RAST server listed closest neighbors based on functional comparison of genome sequences, including *E. coli* AA86 (score 544), *E. coli* 96.0107 (score 284), *Klebsiella* sp. 1_1_55 (score 275), *Klebsiella pneumoniae* 342 (score 274), *Klebsiella oxytoca* 10 to 5,246 (score 250), and *Klebsiella variicola* AT-22 (score 265). RAST predicted 5,462 coding sequences and 101 RNAs representing 572 subsystems. Several genes were identified that were associated with resistance to heavy metals (arsenic, cobalt, zinc, chromium, cadmium, mercury, and copper), as well as resistance to antibiotics [fluoroquinolones, fosfomycin, β-lactamase, multiple antibiotic resistance (mar) locus, the mdtABCD multidrug resistance cluster, and multidrug resistance efflux pumps]. Identification of antimicrobial resistance genes and carbohydrate metabolism of this bacterium will facilitate formulation of novel chromogenic/fluorogenic agar media to support exclusive growth and specific identification of *E. coli*.

**Nucleotide sequence accession numbers.** This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession no. JPRG00000000. The version described in this paper is version JPRG01000000.

**ACKNOWLEDGMENTS**

This work was supported by the Water Research Foundation (WaterRF Project 4238).

The use of trade names and names of commercial sources is for identification only and does not imply endorsement by the CDC or the U.S. Department of Health and Human Services. The findings and conclusions in this report are those of the authors and do not necessarily represent those of the CDC.

**REFERENCES**

1. Brenner KP, Rankin CC, Roybal YR, Stelma GN, Jr, Scarpino PV, Dufour AP. 1993. New medium for the simultaneous detection of total coliforms and *Escherichia coli* in water. Appl. Environ. Microbiol. 59: 3534–3544.

2. Sarhan HR, Foster HA. 1991. A rapid fluorogenic method for the detection of *Escherichia coli* by the production of beta-glucuronidase. J. Appl. Bacteriol. 70:394–400. [http://dx.doi.org/10.1111/j.1365-2672.1991.tb02955.x](http://dx.doi.org/10.1111/j.1365-2672.1991.tb02955.x)

3. Tryland I, Fiksdal L. 1998. Enzyme characteristics of beta-D-
galactosidase- and beta-D-glucuronidase-positive bacteria and their interference in rapid methods for detection of waterborne coliforms and *Escherichia coli*. Appl. Environ. Microbiol. 64:1018–1023.

4. Mollet C, Drancourt M, Raoult D. 1997. rpoB sequence analysis as a novel basis for bacterial identification. Mol. Microbiol. 26:1005–1011. [http://dx.doi.org/10.1046/j.1365-2958.1997.6382009.x](http://dx.doi.org/10.1046/j.1365-2958.1997.6382009.x).

5. Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeck RA, McNeil IK, Paarmann D, Paczian T, Parrello B, Pusch GD, Reich C, Stevens R, Vassieva O, Vonstein V, Wilke A, Zagnitko O. 2008. The RAST server: Rapid Annotations using Subsystems Technology. BMC Genomics 9:75. [http://dx.doi.org/10.1186/1471-2164-9-75](http://dx.doi.org/10.1186/1471-2164-9-75).