AMERICAN SOCIETY FOR MICROBIOLOGY | genomeAnnouncements

Draft Genome Sequences of Two "*Haemophilus quentini*" Isolates Recovered from Two Different Patients' Blood Cultures

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Here, we present the draft genome sequences of two strains (K068 and C860) of the genospecies "*Haemophilus quentini*." The isolates were recovered from blood cultures of a newborn neonate and an elderly patient with septicemia in Ontario, Canada.

Received 30 September 2016 **Accepted** 7 October 2016 **Published** 23 November 2016

Citation Eshaghi A, Soares D, Tsang R, Richardson D, Kus JV, Patel SN. 2016. Draft genome sequences of two "*Haemophilus quentini*" isolates recovered from two different patients' blood cultures. Genome Announc 4(6):e01321-16. doi:10.1128/genomeA.01321-16.

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In humans, *Haemophilus influenzae* strains are often limited to the upper respiratory tract; however, there have been reports of infections in the urogenital tracts. Most of these urogenital infections are caused by nontypeable (NT) *H. influenzae* biotype IV strains (1, 2). Invasive cases of NT *H. influenzae* during the perinatal period have also been reported in the literature (3, 4). Molecular analysis of NT *H. influenzae* biotype IV identified a subset of closely related strains which were initially referred to as "*H. influenzae* cryptic genospecies," or, "*Haemophilus quentini*" (5). *H. quentini* can only be clearly differentiated from *H. influenzae* using 16S rRNA full-gene sequencing, as traditional phenotypic and biochemical tests are not useful in identifying *H. quentini*. Interestingly, 16S rRNA gene sequence is more closely related to *Haemophilus haemolyticus* than *H. influenzae* (6). Here, we report draft genome sequences of two *H. quentini* isolates recovered from blood cultures from two different patients. The biochemical results suggested *H. influenzae*. However, it was ornithine positive. As a result, 16S rRNA sequencing was performed, which suggested *H. haemolyticus*. Due to the discrepant results, the cultures were sent to National Microbiology Laboratory (NML) for *H. quentini*-specific testing. The NML performed *H. quentini*-specific molecular testing to confirm their identification.

Genomic DNA of *H. quentini* isolates K068 and C860 was extracted and purified using a QIAamp DNA minikit (Qiagen, Valencia, CA, USA) from an overnight culture on a blood agar plate. The samples were indexed during library preparation using the Nextera XT DNA sample preparation kit (Illumina, San Diego, CA, USA). The sequencing library was quantified using Qubit 2.0 (Invitrogen, Waltham, MA, USA), and concentration and quality were analyzed by Bioanalyzer (Agilent Technologies, Richardson, TX, USA). The pooled libraries were sequenced using MiSeq Illumina with the V2 kit (2 × 150 bp), according to the manufacturer's instructions, generating 2,735,812 and 1,308,728 high-quality reads corresponding to 407,844,516 and 190,672,282 detected bases for *H. quentini* strains K068 and C860, respectively.

The raw Illumina reads were trimmed and assembled using the *de novo* assembler in CLC Genomics Workbench version 8.0.1 (CLC bio, Germantown, MD, USA). The number of contigs per assembly was 63 and 78 for strains K068 and C860, respectively. All contigs less than 500 bp were filtered, and the remaining 59 contigs (N_{50}, 69,953 bp) (K068) and 71 contigs (N_{50}, 66,205 bp) (C860) were used for annotation. Using the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) (http://www.ncbi.nlm.nih.gov/genome/annotation_prok), 1,989 predicted protein-coding sequences (CDSs), 7 rRNAs, and 48 tRNAs were annotated for *H. quentini* strain K068, while 1,983 CDSs, 5 rRNAs, and 49 tRNAs were annotated for *H. quentini* strain C860.

The functional comparison of the genome sequences available on the Rapid Annotations using Subsystems Technology (RAST) server revealed the closest neighbor of our *H. quentini* isolates to be *H. influenzae* R2866 (score, 507), followed by *H. influenzae* PittGG (score, 500) (7).

Accession number(s). The draft genome sequences for both isolates have been deposited at GenBank under the accession numbers MDJB00000000 (K068) and MDJC00000000 (C860).

ACKNOWLEDGMENTS

We thank Aimin Li and the staff in the DNA core facility and in the reference identification (Abdul Latheef and Hanyue Zhang) section for their excellent technical support and assistance.

We declare no conflicts of interest.

REFERENCES

1. **Wallace RJ, Baker CJ, Quinones FJ, Hollis DG, Weaver RE, Wiss K.** 1983. Nontypable *Haemophilus influenza* (biotype 4) as a neonatal, maternal, and genital pathogen. Rev Infect Dis **5:**126–136.
2. **Quentin R, Martin C, Musser JM, Pasquier-Picard N, Goudeau A.** 1993. Genetic characterization of cryptic genospecies of *Haemophilus* causing urogenital and neonatal infections. J Clin Microbiol **31:** 1111–1116.
3. **Giufrè M, Cardines R, Degl'Innocenti R, Cerquetti M.** 2015. First report of neonatal bacteremia caused by *Haemophilus quentini* diagnosed by 16S rRNA gene sequencing analysis. Diagn Microbiol Infect Dis **83:**121–123. http://dx.doi.org/10.1016/j.diagmicrobio.2015.05.019.
4. **Collins S, Litt DJ, Flynn S, Ramsay ME, Slack MP, Ladhani SN.** 2015. Neonatal invasive *Haemophilus influenzae* disease in England and Wales:

epidemiology, clinical characteristics, and outcome. Clin Infect Dis **60:** 1786–1792. http://dx.doi.org/10.1093/cid/civ194.

5. **Quentin R, Ruimy R, Rosenau A, Musser JM, Christen R**. 1996. Genetic identification of cryptic genospecies of *Haemophilus* causing urogenital and neonatal infections by PCR using specific primers targeting genes coding for 16S rRNA. J Clin Microbiol **34:**1380–1385.
6. **Glover WA, Suarez CJ, Clarridge JE**. 2011. Genotypic and phenotypic characterization and clinical significance of *Haemophilus quentini* isolated from the urinary tract of adult men. J Med Microbiol **60:**1689–1692. http://dx.doi.org/10.1099/jmm.0.031591-0.
7. **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, Overbeek RA, McNeil LK, 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.