Draft Genome Sequence of the Earliest *Cronobacter sakazakii* Sequence Type 4 Strain, NCIMB 8272

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The *Cronobacter sakazakii* clonal lineage defined as sequence type 4 (ST4) is associated with severe cases of neonatal meningitis and persistence in powdered infant formula. For genome sequencing of the earliest deposited culture collection strain of *Cronobacter sakazakii* ST4, we used the strain NCIMB 8272, originally isolated from milk powder in 1950.

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he Cronobacter genus is associated with severe human infections such as meningitis, septicemia, and necrotizing enterocolitis (1). A multilocus sequence typing scheme has been established for the genus and over 200 sequence types (ST) have been defined (1). Previous studies of Cronobacter isolates collected over a 35-year period across 6 countries have revealed a strong association of Cronobacter sakazakii sequence type 4 (ST4) with neonatal meningitis (2). In addition, C. sakazakii ST4 has frequently (up to 24% of strains) been isolated from powdered infant formula (PIF) manufacturing plants as well as from PIF (3, 4). Therefore, efforts to improve understanding of C. sakazakii ST4 are warranted to gain insight regarding its prevalence in manufacturing plants and infant formula and its pathogenicity. In this study, we sequenced C. sakazakii NCIMB 8272 as it is the oldest strain in international culture collections, having been isolated from milk powder in 1950 (1). This strain can thus be compared with more recently isolated and sequenced Cronobacter species (5).

C. sakazakii NCIMB 8272 DNA was extracted from 1-day cultures using a GenElute bacterial genome kit (Sigma-Aldrich) and sequenced using an Illumina HiSeq2500 system. A total of 7,102,990 high-quality paired-end reads 150 bp in length, with 30-fold coverage, were generated. *De novo* assembly was performed using Velvet (6). Further annotation was performed using the SEED-based automated annotation system provided by the RAST Server (7).

The genome sequence of *C. sakazakii* NCIMB 8272 was 4,583,199 bp in length with a G+C content of 56.8%. The draft genome was distributed in 104 contigs with 4,259 coding sequences (CDS) and 97 RNAs.

The CDS included genes associated with iron acquisition, stress response, heavy metal resistance (arsenic, silver cobalt, zinc, and cadmium), and phages. Several virulence-associated traits such as adhesion and sialic acid utilization were also determined. These have previously been described in *Cronobacter* (5).

Nucleotide sequence accession number. The genome sequences of *C. sakazakii* NCIMB 8272 have been deposited in GenBank under the accession number AWFW00000000.

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REFERENCES

- 1. Joseph S, Sonbol H, Hariri S, Desai P, McClelland M, Forsythe SJ. 2012. Diversity of the *Cronobacter* genus as revealed by multilocus sequence typing. J. Clin. Microbiol. 50:3031–3039.
- Hariri S, Joseph S, Forsythe SJ. 2013. Predominance of *Cronobacter saka-zakii* ST4 clonal complex strains in *Cronobacter* neonatal meningitis infections in US 2011. Emerg. Infect. Dis. 19:175–177.
- Müller A, Stephan R, Fricker-Feer C, Lehner A. 2013. Genetic diversity of *Cronobacter sakazakii* isolates collected from a Swiss infant formula production facility. J. Food Prot. 76:883–887.
- 4. Sonbol H, Joseph S, McAuley C, Craven H, Forsythe SJ. 2013. Multilocus sequence typing of *Cronobacter* spp. from powdered infant formula and milk powder production factories. Int. Dairy J. **30**:1–7.
- Joseph S, Desai P, Ji Y, Cummings CA, Shih R, Degoricija L, Rico A, Brzoska P, Hamby SE, Masood N, Hariri S, Sonbol H, Chuzhanova N, McClelland M, Furtado MR, Forsythe SJ. 2012. Comparative analysis of genome sequences covering the seven *Cronobacter* species. PLOS One 7:e49455.
- 6. Zerbino DR, Birney E. 2008. Velvet: algorithms for *de novo* short read assembly using de Bruijn graphs. Genome Res. **18**:821–829.
- 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. doi:10.1186/1471-2164-9-75.