

Complete Genome and Plasmid Sequences of Three Canadian Strains of *Salmonella enterica* subsp. *enterica* Serovar Enteritidis Belonging to Phage Types 8, 13, and 13a

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***Salmonella enterica* subsp. *enterica* serovar Enteritidis is a prominent cause of human salmonellosis frequently linked to poultry products. In Canada, *S. Enteritidis* phage types 8, 13, and 13a predominate among both clinical and poultry isolates. Here, we report the complete genome and plasmid sequences of poultry isolates of these three phage types.**

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Nontyphoidal salmonellosis is one of the most common zoonotic foodborne infections worldwide (1). Over the past decade, *Salmonella enterica* subsp. *enterica* serovar Enteritidis has become the predominant serovar isolated from humans in Canada, accounting annually for 30 to 40% of all reported clinical isolates (2–4). Phage typing of *S. Enteritidis* (5) has revealed geographic differences in the predominant phage types (PTs). In Europe, PTs 4, 1, 8, and 21 are the most common (6), whereas in Canada, ~60% of human isolates are PTs 8, 13, and 13a (3, 4). The same three PTs predominate among nonhuman *S. Enteritidis* isolates submitted to our World Organisation for Animal Health (OIE) Reference Laboratory for Salmonellosis, of which ~85% are from chickens or chicken products. Here, we announce the availability of the complete genome and plasmid sequences of one isolate each of PT 8, 13, and 13a from poultry sources in Canada that may serve as reference genomes for comparative analyses.

S. Enteritidis poultry isolates H10-082405-11 (PT8), SA01AB08065501 (PT13), and SA01AB10104801 (PT13a) were propagated in tryptic soy broth. Genomic DNA was prepared by using the Qiagen EZ1 DNA tissue kit. Sequencing was performed on two platforms: Pacific Biosciences (PacBio) RS II (Menlo Park, CA, USA), and Illumina MiSeq (Illumina, Inc., San Diego, CA). PacBio (performed at the McGill University and Génome Québec Innovation Centre, Montréal, Québec, Canada) generated

326,964 raw subreads of average length 2,885 to 3,240 bp using two single-molecule real-time (SMRT) cells. The contigs had an average coverage of 10.65× to 22.88× and were assembled *de novo* using Hierarchical Genome Assembly Process (HGAP) (7). Sequencing on the Illumina MiSeq platform was performed at the Public Health Agency of Canada (PHAC) National Microbiology Laboratory, Winnipeg, Canada, with 101 × 2 paired-end reads with an average 84.2× coverage after library preparation, using the Illumina Nextera XT kit. The Illumina reads were analyzed and quality checked using FastQC (<http://www.bioinformatics.babraham.ac.uk/projects/fastqc/>). An *in silico* *NcoI* restriction map of the GenBank *S. Enteritidis* PT4 strain P125109 (GenBank accession no. AM933172) was generated and used to verify contig assembly using MapSolver version 2.1.1 (OpGen, Inc., Madison, WI). Genome assemblies were created by using the MIRA assembler version 4.9.3 (8) and by manually checking potential joins using the Gap5 software of the Staden package (9). Comparison with the *in silico* restriction map of GenBank strain P125109 and with closely related plasmid sequences in GenBank, together with the finishing process, produced fully assembled genomes and plasmids. The genomes consisted of single chromosomal contigs ranging from 4,679,291 to 4,712,309 bp, with an average G+C content of ~52.17%. The plasmid contigs ranged from 34,541 to 59,372 bp, with G+C content ranging from ~41.45 to 51.96%.

TABLE 1 Accession and isolate numbers for the three *Salmonella* Enteritidis genomes and plasmids sequenced in this study

GenBank accession no.	Isolate accession no.	Original isolate no.	Phage type	PFGE type ^a
CP007329 KT317612	EC20120002 pSE_EC20120002_60	H10-082405-11	8	SENXAI.0003, SENBNI.0003
CP007249 KT317611	EC20090641 pSE_EC20090641_60	SA01AB08065501	13	SENXAI.0038, SENBNI.0016
CP007267 KT317613 KT317614	EC20120005 pSE_EC20120005_60 pSE_EC20120005_35	SA01AB10104801	13a	SENXAI.0006, SENBNI.0007

^a PFGE, pulsed-field gel electrophoresis.

The genomes and plasmids were annotated using the National Center for Biotechnology Information (NCBI) Prokaryotic Genomes Annotation Pipeline (PGAP) (<http://ncbi.nlm.nih.gov/genomes/static/Pipeline.html>), identifying ~4,447 to 4,486 coding DNA sequences (CDSs) per genome and ~43 to 74 CDSs per plasmid.

Nucleotide sequence accession numbers. The complete genome and plasmid sequences of these three *S. Enteritidis* strains have been deposited in GenBank under BioProject no. 219482. The GenBank accession numbers are listed in [Table 1](#).

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REFERENCES

1. Majowicz SE, Musto J, Scallan E, Angulo FJ, Kirk M, O'Brien SJ, Jones TF, Fazil A, Hoekstra RM, International Collaboration on Enteric Disease 'Burden of Illness' Studies. 2010. The global burden of nontyphoidal *Salmonella* gastroenteritis. *Clin Infect Dis* 50:882–889. <http://dx.doi.org/10.1086/650733>.
2. Nesbitt A, Ravel A, Murray R, McCormick R, Savelli C, Finley R, Parmley J, Agunos A, Majowicz SE, Gilmour M, Canadian Integrated Program for Antimicrobial Resistance Surveillance Public Health Partnership, Canadian Public Health Laboratory Network. 2012. Integrated surveillance and potential sources of *Salmonella* Enteritidis in human cases in Canada from 2003 to 2009. *Epidemiol Infect* 140:1757–1772. <http://dx.doi.org/10.1017/S0950268811002548>.
3. Landry L, Dutil L. 2010. Epidemiology of SE in humans in Canada, Canadian *Salmonella* Enteritidis Control Symposium, 1 December 2010, Vancouver, British Columbia, Canada.
4. Public Health Agency of Canada. 2015. The National Enteric Surveillance Program (NESP) Annual summary 2013. Public Health Agency of Canada, Winnipeg, Manitoba, Canada.
5. Ward LR, de Sa JD, Rowe B. 1987. A phage-typing scheme for *Salmonella* Enteritidis. *Epidemiol Infect* 99:291–294. <http://dx.doi.org/10.1017/S095026880067765>.
6. Peters TM, Berghold C, Brown D, Coia J, Dionisi AM, Echeita A, Fisher IS, Gatto AJ, Gill N, Green J, Gerner-Smidt P, Heck M, Lederer I, Lukinmaa S, Luzzi I, Maguire C, Prager R, Usera M, Siitonen A, Threlfall EJ, Torpdahl M, Tschape H, Wannet W, Zwaluw WK. 2007. Relationship of pulsed-field profiles with key phage types of *Salmonella enterica* serotype Enteritidis in Europe: results of an international multi-centre study. *Epidemiol Infect* 135:1274–1281. <http://dx.doi.org/10.1017/S0950268807008102>.
7. Chin CS, Alexander DH, Marks P, Klammer AA, Drake J, Heiner C, Clum A, Copeland A, Huddleston J, Eichler EE, Turner SW, Korlach J. 2013. Nonhybrid, finished microbial genome assemblies from long-read SMRT sequencing data. *Nat Methods* 10:563–569. <http://dx.doi.org/10.1038/nmeth.2474>.
8. Chevreur B, Wetter T, Suhai S. 1999. Genome sequence assembly using trace signals and additional sequence information, p 45–56. *In* Computer science and biology. Proceedings of the German Conference on Bioinformatics, GCB '99. GCB, Hannover, Germany.
9. Staden R, Beal KF, Bonfield JK. 2000. The Staden package, 1998. *Methods Mol Biol* 132:115–130.