

Complete Genome Sequence of Universal Bacteriophage Host Strain Campylobacter jejuni subsp. jejuni PT14

Kelly J. Brathwaite,^a Patcharin Siringan,^a Joanna Moreton,^b Ray Wilson,^b Ian F. Connerton^a

Division of Food Sciences, University of Nottingham, Sutton Bonington Campus, Loughborough, United Kingdom^a; Deep Seq, Centre for Genetics and Genomics, University of Nottingham, Queen's Medical Center, Nottingham, United Kingdom^b

Campylobacter jejuni strain PT14 is a clinical isolate previously used to propagate bacteriophages in the United Kingdom phage typing scheme. The strain has proven useful in the isolation of *Campylobacter* bacteriophages from several sources, and it functions as a model host in phage therapy experiments with poultry and poultry meat.

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Address correspondence to Ian F. Connerton, ian.connerton@nottingham.ac.uk.

Campylobacter jejuni PT14 has been used to isolate and propagate bacteriophages from environmental samples (1–6). The strain is available from the Public Health England Board as NCTC 12662 (http://www.phe-culturecollections.org.uk). The genome sequence of *C. jejuni* PT14 was determined by pyrosequencing on a 454 GS FLX platform (Roche Diagnostics). A total of 257,645 reads were generated, with an average read length of 352 bases. The reads were *de novo* assembled into a single contig using the CLC Genomics Workbench 6.0. The sequence was compared and confirmed with 5 million 50-bp reads generated using MiSeq technology operating in paired-end mode (Illumina). The genome sequence was annotated using the Prokaryotic Genome Automatic Annotation Pipeline (PGAAP) (7).

The circular genome of *C. jejuni* PT14 was found to be 1,635,252 bp in length, with 1,607 coding sequences and an average G+C content of 30.5%. The genome sequence contains 3 rRNA operons and 41 tRNA genes. No prophage-associated genes or plasmids were found in this genome. *C. jejuni* PT14 contains 26 probable pseudogenes, but notably, it has three annotated pseudogenes from *C. jejuni* strain NCTC 11168, Cj0501, Cj1064, and Cj1470c, which remain intact within the PT14 genome. The clustered regularly interspaced short palindromic repeats and associated genes (CRISPR-Cas) within the genome of *C. jejuni* PT14 were found to be intact, which is an interesting finding considering the sensitive nature of this strain to bacteriophage infections. The CRISPR element is comprised of 32-bp-long spacer sequences and four repeat regions.

C. jejuni PT14 was found to contain 27 homopolymeric G+C tracts (defined as containing \geq 7 consecutive G+C residues), which bears comparison to the 29 reported in *C. jejuni* NCTC 11168, 25 in RM1221, and 23 in CG8486 (8–11). The locations of the homopolymeric tracts are generally conserved among the *C. jejuni* strains. In *C. jejuni* PT14, there are five tracts residing within intergenic regions, two in probable pseudogenes, and nine within genes of unknown function. Variation in the length of the G+C tracts within genes results in phase-variable expression. Five of the tracts identified in *C. jejuni* PT14, including genes encoding

two putative methyltransferases and the invasion protein CipA, show phase variation at the sequence level. C. jejuni PT14 also contains a phase-variable A+T region in the gene A911_06060, which encodes a GMP synthase. Phase-variable gene expression has been correlated with modifications of C. jejuni surface structures that are required for bacteriophage infection (12). Phasevariable disruption of the Cj1421 pseudogene of C. jejuni NCTC 11168 prevents O-methyl phosphoramidate attachment to GalfNAc of a capsular polysaccharide, which leads to noninfection by bacteriophage F336 (13) and allows bacteriophage evasion during chicken colonization (14). The Cj1421 homologue of C. jejuni PT14 (A911_06918) does not appear in a syntenic region and is not phase variable. However, the Cj1422 homologue (A911_06907), which hinders phage infection by O-methyl phosphoamidate attachment to heptose, exhibits phase variation. In summary, although C. jejuni PT14 is a phage-sensitive strain, bacterial defense mechanisms still appear to be in place to enable its escape from bacteriophage predation.

Nucleotide sequence accession number. The *C. jejuni* PT14 sequence is available under GenBank accession no. CP003871.

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REFERENCES

- Frost JA, Kramer JM, Gillanders SA. 1999. Phage typing of *Campylobacter jejuni* and *Campylobacter coli* and its use as an adjunct to serotyping. Epidemiol. Infect. 123:47–55.
- Atterbury RJ, Connerton PL, Dodd CE, Rees CE, Connerton IF. 2003. Isolation and characterization of *Campylobacter* bacteriophages from retail poultry. Appl. Environ. Microbiol. 69:4511–4518.
- Atterbury RJ, Connerton PL, Dodd CE, Rees CE, Connerton IF. 2003. Application of host-specific bacteriophages to the surface of chicken skin leads to a reduction in recovery of *Campylobacter jejuni*. Appl. Environ. Microbiol. 69:6302–6306.
- Connerton PL, Loc Carrillo CM, Swift C, Dillon E, Scott A, Rees CE, Dodd CE, Frost J, Connerton IF. 2004. Longitudinal study of *Campylo*-

bacter jejuni bacteriophages and their hosts from broiler chickens. Appl. Environ. Microbiol. **70:**3877–3883.

- El-Shibiny A, Connerton PL, Connerton IF. 2005. Enumeration and diversity of campylobacters and bacteriophages isolated during the rearing cycles of free-range and organic chickens. Appl. Environ. Microbiol. 71:1259–1266.
- Loc Carrillo C, Atterbury RJ, El-Shibiny A, Connerton PL, Dillon E, Scott A, Connerton IF. 2005. Bacteriophage therapy to reduce *Campylobacter jejuni* colonization of broiler chickens. Appl. Environ. Microbiol. 71:6554–6563.
- Angiuoli SV, Gussman A, Klimke W, Cochrane G, Field D, Garrity G, Kodira CD, Kyrpides N, Madupu R, Markowitz V, Tatusova T, Thomson N, White O. 2008. Toward an online repository of Standard Operating Procedures (SOPs) for (meta)genomic annotation. Omics 12:137–141.
- 8. Gundogdu O, Bentley SD, Holden MT, Parkhill J, Dorrell N, Wren BW. 2007. Re-annotation and re-analysis of the *Campylobacter jejuni* NCTC11168 genome sequence. BMC Genomics 8:162. doi:10.1186/1471 -2164-8-162.
- Parker CT, Quiñones B, Miller WG, Horn ST, Mandrell RE. 2006. Comparative genomic analysis of *Campylobacter jejuni* strains reveals diversity due to genomic elements similar to those present in *C. jejuni* strain RM1221. J. Clin. Microbiol. 44:4125–4135.

- Parkhill J, Wren BW, Mungall K, Ketley JM, Churcher C, Basham D, Chillingworth T, Davies RM, Feltwell T, Holroyd S, Jagels K, Karlyshev AV, Moule S, Pallen MJ, Penn CW, Quail MA, Rajandream MA, Rutherford KM, van Vliet AH, Whitehead S, Barrell BG. 2000. The genome sequence of the food-borne pathogen *Campylobacter jejuni* reveals hypervariable sequences. Nature 403:665–668.
- 11. Poly F, Read T, Tribble DR, Baqar S, Lorenzo M, Guerry P. 2007. Genome sequence of a clinical isolate of *Campylobacter jejuni* from Thailand. Infect. Immun. **75**:3425–3433.
- 12. Coward C, Grant AJ, Swift C, Philp J, Towler R, Heydarian M, Frost JA, Maskell DJ. 2006. Phase-variable surface structures are required for infection of *Campylobacter jejuni* by bacteriophages. Appl. Environ. Microbiol. 72:4638–4647.
- Sørensen MC, van Alphen LB, Harboe A, Li J, Christensen BB, Szymanski CM, Brøndsted L. 2011. Bacteriophage F336 recognizes the capsular phosphoramidate modification of *Campylobacter jejuni* NCTC 11168. J. Bacteriol. 193:6742–6749.
- Sørensen MCH, van Alphen LB, Fodor C, Crowley SM, Christensen BB, Szymanski CM, Brøndsted L. 2012. Phase variable expression of capsular polysaccharide modifications allows *Campylobacter jejuni* to avoid bacteriophage infections in chickens. Front. Cell. Infect. Microbiol. 2:1–11.