

Genome Sequence of *Chlamydia psittaci* Strain 01DC12 Originating from Swine

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Chlamydia psittaci is the etiological agent of psittacosis and is a zoonotic pathogen infecting birds and a variety of mammalian hosts. Here we report the genome sequence of the porcine strain 01DC12 which is representative of a novel clade of *C. psittaci* belonging to *ompA* genotype E.

Received 30 October 2012 Accepted 7 November 2012 Published 15 January 2013

Citation Seth-Smith HMB, Sait M, Sachse K, Gaede W, Longbottom D, Thomson NR. 2013. Genome sequence of *Chlamydia psittaci* strain 01DC12 originating from swine. Genome Announc. 1(1):e00078-12. doi:10.1128/genomeA.00078-12.

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Chlamydia psittaci is a Gram-negative, obligate intracellular bacterium and is the etiological agent of psittacosis, causing disease primarily in birds but also infecting a variety of mammals, including humans. *C. psittaci* has been isolated from the lungs (1), genital tract (2), and semen (3) of pigs. At least 15 different genotypes have been defined on the basis of the *ompA* gene sequence variation (4). Here we report the genome sequence of 01DC12, a strain that was isolated from a pig in Saxony-Anhalt, Germany.

The C. psittaci 01DC12 genome was sequenced using the Illumina HiSeq platform with 75-bp paired-end reads, resulting in a mean genome coverage of 248×. Reads were assembled using Velvet v1.0.12 (5) to produce 8 contigs, which were ordered against the C. psittaci strain RD1 genome (6). These were finished manually and using GapFiller (7), resulting in an improved high-quality draft genome sequence (8) consisting of 2 contigs. The chromosome of C. psittaci 01DC12 comprises 1,171,011 bp, with G+C content of 39.0%. Annotation was transferred from strain RD1 using annotations_update (https://github.com/sanger-pathogens /annotations_update) on the basis of BLASTN similarity using the default settings, and manually curated using Artemis (9). Comparative analysis with closely related C. psittaci strains (10-12) was performed using ACT (13). The remaining gap, estimated at 2599 bp, is predicted to encode C and N termini of repetitive pmp genes. This region plus the gaps filled using GapFiller are marked in the genome annotation.

The genome contains 963 predicted coding sequences (CDSs), a single rRNA operon, and 38 tRNA genes. Analysis of the *ompA* gene confirms that *C. psittaci* 01DC12 belongs to genotype E. Sequence analysis of the 7 loci used for multilocus sequence typing (MLST) (14) assigned *gatA*, *hemN*, and *hflX* to allele 11, *enoA* and *fumC* to allele 13, and *gidA* and *oppA* to allele 14, indicating that *C. psittaci* 01DC12 is a novel MLST sequence type. Comparison with the genome of type strain 6BC (10, 11) shows that the 01DC12 chromosome contains deletions of 360 bp and 144 bp, respectively, in two putative membrane proteins (BN356_2661

and BN356_4221) and a deletion of an 802-bp repeat at the C terminus of an incA/TMH-family membrane protein (BN356_7881). Comparative mapping against strain 6BC revealed 6,262 single-nucleotide polymorphisms (SNPs), suggesting that strain 01DC12 represents a new lineage within C. psittaci. A total of 11 pseudogenes were identified in the genome of 01DC12, including those predicted to encode an aldolase (BN356_2651), Pmp8G (BN356 2811), 6 membrane proteins (BN356 3711, BN356_5531, BN356_5541, BN356_5601, BN356_5741, and BN356_6271), a Mycobacterium avium complex (MAC)/perforindomain protein (BN356_5611), and an IncA-family protein (BN356_7871). The genome of strain 01DC12 includes a single plasmid of 7553 bp with G+C content of 32.8%, predicted to encode 8 CDSs. Plasmid p01DC12 is differentiated from the plasmid carried by strain 6BC by 2 SNPs (10, 11): one synonymous SNP in a putative helicase (BN356_p003) and one nonsynonymous SNP resulting in a T-I change in hypothetical protein BN356_p008.

Nucleotide sequence accession numbers. The genome and plasmid sequences of *C. psittaci* 01DC12 have been deposited in EMBL under accession numbers HF545614 and HF545615.

ACKNOWLEDGMENTS

This work was supported by the Wellcome Trust (grant number 098051), by the Biotechnology and Biological Sciences Research Council (BBSRC) (grant number BB/E018939/1), and by the Scottish Government Rural and Environment Research and Analysis Directorate (RERAD).

REFERENCES

- Vanrompay D, Geens T, Desplanques A, Hoang TQ, De Vos L, Van Loock M, Huyck E, Cox E, Cox E. 2004. Immunoblotting, ELISA and culture evidence for Chlamydiaceae in sows on 258 Belgian farms. Vet. Microbiol. 99(1):56–66.
- Busch M, Thoma R, Schiller I, Corboz L, Pospischil A. 2000. Occurrence of chlamydiae in the genital tracts of sows at slaughter and their possible significance for reproductive failure. J. Vet. Med. B Infect. Dis. Vet. Public Health 47(6):471–480.

- Kauffold J, Melzer F, Henning K, Schulze K, Leiding C, Sachse K. 2006. Prevalence of chlamydiae in boars and semen used for artificial insemination. Theriogenology 65(9):1750–1758.
- Sachse K, Laroucau K, Hotzel H, Schubert E, Ehricht R, Slickers P. 2008. Genotyping of *Chlamydophila psittaci* using a new DNA microarray assay based on sequence analysis of *ompA* genes. BMC Microbiol. 8:63.
- Zerbino DR, Birney E. 2008. Velvet: algorithms for de novo short read assembly using de Bruijn graphs. Genome Res. 18(5):821–829.
- Seth-Smith HM, Harris SR, Range R, West AP, Severin JA, Ossewaarde JM, Cutcliffe LT, Skilton RJ, Marsh P, Parkhill J, Clarke IN, Thomson NR. 2011. Genome sequence of the zoonotic pathogen *Chlamydophila psittaci*. J. Bacteriol. 193(5):1282–1283.
- 7. Boetzer M, Pirovano W. 2012. Toward almost closed genomes with GapFiller. Genome Biol. 13(6):R56.
- 8. Chain PSG, Grafham DV, Fulton RS, FitzGerald MG, Hostetler J, Muzny D, Ali J, Birren B, Bruce DC, Buhay C, Cole JR, Ding Y, Dugan S, Field D, Garrity GM, Gibbs R, Graves T, Han S, Harrison SH, Highlander S, Hugenholtz P, Khouri HM, Kodira CD, Kolker E, Kyrpides NC, Lang D, Lapidus A, Malfatti SA, Markowitz V, Metha T, Nelson KEKE, Parkhill J, Pitluck S, Qin X, Read TD, Schmutz J, Sozhamannan S, Sterk P, Strausberg RL, Sutton G, Thomson NR, Tiedje JM, Weinstock G, Wollam A, Genomic Standards Consortium Human Microbiome Project Jumpstart Consortium, Detter JC. 2009.

Genome project standards in a new era of sequencing. Science 326: 236-237.

- 9. Rutherford K, Parkhill J, Crook J, Horsnell T, Rice P, Rajandream MA, Barrell B. 2000. Artemis: sequence visualization and annotation. Bioinformatics 16(10):944–945.
- Voigt A, Schöfl G, Heidrich A, Sachse K, Saluz HP. 2011. Full-length de novo sequence of the *Chlamydophila psittaci* type strain 6 BC. J. Bacteriol. 193(10):2662–2663.
- Grinblat-Huse V, Drabek EF, Huot Creasy H, Daugherty SC, Jones KM, Santana-Cruz I, Tallon LJ, Read TD, Hatch TP, Bavoil P, Myers GSA. 2011. Genome sequences of the zoonotic pathogens *Chlamydia psittaci* 6 BC and Cal10. J. Bacteriol. 193(15):4039–4040.
- Schöfl G, Voigt A, Litsche K, Sachse K, Saluz HP. 2011. Complete genome sequences of four mammalian isolates of *Chlamydophila psittaci*. J. Bacteriol. 193(16):4258.
- Carver TJ, Rutherford KM, Berriman M, Rajandream MA, Barrell BG, Parkhill J. 2005. ACT: the Artemis comparison tool. Bioinformatics 21(16):3422–3423.
- 14. Pannekoek Y, Dickx V, Beeckman DSA, Jolley KA, Keijzers WC, Vretou E, Maiden MCJ, Vanrompay D, van der Ende A. 2010. Multi locus sequence typing of chlamydia reveals an association between *Chlamydia psittaci* genotypes and host species. PLoS One 5(12):e14179.