



Genome Sequence of *Coxiella burnetii* Strain AuQ01 (Arandale) from an Australian Patient with Acute Q Fever

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Coxiella burnetii strain AuQ01 was isolated from the serum of an Australian acute Q fever patient and represents the first whole genome from this historical Q fever country. This new genome shows distinct differences from existing genomic data and will enhance the understanding of this query pathogen.

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coxiella burnetii, a Gram-negative coccobacillus belonging to the class *Gammaproteobacteria*, is the causative agent of the zoonosis Q fever. It has a worldwide distribution but the disease is rarely reported, a situation that probably results from the diagnostic challenges presented by *C. burnetii* and lack of requirement for notification of cases in many countries (1). Despite this, Q fever persists with a large reservoir among multiple species, regularly leading to smaller outbreaks.

Since the first publication of the complete genome sequence of the historical Nine Mile strain (2, 3) in 2003, more isolates have been sequenced and published (4–6). Therefore, efforts to enlarge genomic knowledge are important to achieve more information about this pathogen. Although Australia was also the first country in which the Q fever agent was discovered (7), no genome sequence has been available from this continent until now.

Here, we present a strain, initially named Arandale, which was isolated in a cell culture in 2005 from the buffy coat (EDTA blood sample) of a 56-year-old male patient with acute Q fever who lived in Deepwater, New South Wales, Australia (8). Genomic DNA was extracted using MagNA Pure compact system (Roche Diagnostics, Mannheim, Germany). A sequencing library was prepared, with an adapted NEBNext protocol for small amounts of input DNA, and was sequenced by GATC (Constance, Germany) on an Illumina HiSeq 2500 instrument with 2×100 paired-end reads.

Sequence reads were assembled using SPAdes (9). The majority of the gaps in the resulting assembly correspond to the positions of insertion sequence (IS) elements, mostly of type IS1111. IMAGE (10) was used to extend the contigs at the IS ends and Mauve (11) to order them using *C. burnetii* CbuK_Q154 as the reference genome (NCBI reference sequence no. NC_011528 and NC_011526). Some curation and scaffolding at known conserved IS sites (IS30 and ISAs1) and the duplicated region in the plasmid

was done manually. Polishing was done using mapping with Mira (12) followed by variation analysis. Gene calling and functional annotation was performed using the PEDANT system (13).

The *C. burnetii* AuQ01 genome assembly consists of 66 chromosomal contigs of about 2,073,000 bp and a QpRS plasmid of 39,269 bp. The numbers of nearly full-length IS elements were determined to be 55 of type IS1111, 5 of type IS30, and 3 of type ISAs1.

A whole-genome comparison with all five complete reference genomes revealed strain Q154 as the most similar strain. *In silico* typing of strain AuQ01 showed the same *adaA* deletion variant 1 (14), but multispacer sequence typing (MST) and multiple-locus variable number of tandem repeat (VNTR) analysis (MLVA) generated profiles different from those for Q154 (15).

This short comparison demonstrates that *C. burnetii* has a distinct and different evolution on the Australian continent. Furthermore, this new whole-genome sequence enhances the existing genomic data for *C. burnetii* and is therefore beneficial for genomic studies and analyses in the future.

Nucleotide sequence accession number. The whole-genome shotgun project of *C. burnetii* AuQ01 has been deposited at DDBJ/EMBL/GenBank under the accession number JPVV00000000.

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REFERENCES

- Fournier PE, Marrie TJ, Raoult D. 1998. Diagnosis of Q fever. J. Clin. Microbiol. 36:1823–1834.
- Davis GE, Cox HR. 1938. A filter-passing infectious agent isolated from ticks. I. Isolation from *Dermacentor andersoni*, reactions in animals, and

- filtration experiments. Public Health Rep. 53:2259-2261. http://dx.doi.org/10.2307/4582746.
- Seshadri R, Paulsen IT, Eisen JA, Read TD, Nelson KE, Nelson WC, Ward NL, Tettelin H, Davidsen TM, Beanan MJ, Deboy RT, Daugherty SC, Brinkac LM, Madupu R, Dodson RJ, Khouri HM, Lee KH, Carty HA, Scanlan D, Heinzen RA, Thompson HA, Samuel JE, Fraser CM, Heidelberg JF. 2003. Complete genome sequence of the Q-fever pathogen Coxiella burnetii. Proc. Natl. Acad. Sci. U. S. A. 100:5455–5460. http:// dx.doi.org/10.1073/pnas.0931379100.
- Beare PA, Unsworth N, Andoh M, Voth DE, Omsland A, Gilk SD, Williams KP, Sobral BW, Kupko JJ, Porcella SF, Samuel JE, Heinzen RA. 2009. Comparative genomics reveal extensive transposon-mediated genomic plasticity and diversity among potential effector proteins within the genus *Coxiella*. Infect. Immun. 77:642–656. http://dx.doi.org/ 10.1128/IAI.01141-08.
- Karlsson E, Macellaro A, Byström M, Forsman M, Frangoulidis D, Janse I, Larsson P, Lindgren P, Ohrman C, van Rotterdam B, Sjödin A, Myrtennäs K. 2014. Eight new genomes and synthetic controls increase the accessibility of rapid melt-MAMA SNP typing of *Coxiella burnetii*. PLoS One 9:e85417. http://dx.doi.org/10.1371/journal.pone.0085417.
- Sidi-Boumedine K, Ellis RJ, Adam G, Prigent M, Angen O, Aspán A, Thiéry R, Rousset E. 2014. Draft genome sequences of six ruminant Coxiella burnetii isolates of European origin. Genome Announc. 2(3): e00285-14. http://dx.doi.org/10.1128/genomeA.00285-14.
- Burnet FM, Freeman M. 1937. Experimental studies on the virus of "Q" Fever. Med. J. Aust. 2:299–305.
- Lockhart M. 2010. The detection of Coxiella burnetii (Q fever) in clinical and environmental samples. Ph.D. thesis. Murdoch University, Perth, Western Australia.
- 9. Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov

- AS, Lesin VM, Nikolenko SI, Pham S, Prjibelski AD, Pyshkin AV, Sirotkin AV, Vyahhi N, Tesler G, Alekseyev MA, Pevzner PA. 2012. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. J. Comput. Biol. 19:455–477. http://dx.doi.org/10.1089/cmb.2012.0021.
- 10. Tsai IJ, Otto TD, Berriman M. 2010. Improving draft assemblies by iterative mapping and assembly of short reads to eliminate gaps. Genome Biol. 11:R41. http://dx.doi.org/10.1186/gb-2010-11-4-r41.
- Rissman AI, Mau B, Biehl BS, Darling AE, Glasner JD, Perna NT. 2009. Reordering contigs of draft genomes using the Mauve aligner. Bioinformatics 25:2071–2073. http://dx.doi.org/10.1093/bioinformatics/btp356.
- 12. Chevreux B, Wetter T, Suhai S. 1999. Genome sequence assembly using trace signals and additional sequence Information. German Conference on Bioinformatics. Hannover, Germany.
- Walter MC, Rattei T, Arnold R, Güldener U, Münsterkötter M, Nenova K, Kastenmüller G, Tischler P, Wölling A, Volz A, Pongratz N, Jost R, Mewes HW, Frishman D. 2009. PEDANT covers all complete RefSeq genomes. Nucleic Acids Res. 37:D408–D411. http://dx.doi.org/10.1093/ nar/gkn749.
- 14. Frangoulidis D, Splettstoesser WD, Landt O, Dehnhardt J, Henning K, Hilbert A, Bauer T, Antwerpen M, Meyer H, Walter MC, Knobloch JK. 2013. Microevolution of the chromosomal region of acute disease antigen A (*adaA*) in the query (Q) fever agent *Coxiella burnetii*. PLoS One 8:e53440. http://dx.doi.org/10.1371/journal.pone.0053440.
- Frangoulidis D, Walter MC, Antwerpen M, Zimmermann P, Janowetz B, Alex M, Böttcher J, Henning K, Hilbert A, Ganter M, Runge M, Münsterkötter M, Splettstoesser WD, Hanczaruk M. 27 June 2014. Molecular analysis of *Coxiella burnetii* in Germany reveals evolution of unique clonal clusters. Int. J. Med. Microbiol. http://dx.doi.org/10.1016/ j.ijmm.2014.06.011.