



Draft Genome Sequence of *Dichelobacter nodosus* ATCC 25549, Strain VPI 2340 [11342], a Bacterium Causing Footrot in Sheep

Alexandra Jackson,^a María Victoria Humbert,^a Anish Pandey,^b Holly Bratcher,^c Myron Christodoulides^a

Neisseria Research Laboratory^a and Molecular Microbiology,^b Academic Unit of Clinical and Experimental Sciences, Sir Henry Wellcome Laboratories, University of Southampton Faculty of Medicine, Southampton, United Kingdom; Department of Zoology, University of Oxford, Oxford, United Kingdom^c

We report a draft genome sequence for *Dichelobacter nodosus* ATCC 25549, strain VPI 2340 [11342], a causative agent of ovine footrot. The draft genome shares ~98% gene similarity with the available genome of *D. nodosus* strain VCS1703A but is differentiated by extensive gene duplication and the absence of 13 particular genes.

Received 2 August 2015 Accepted 11 August 2015 Published 24 September 2015

Citation Jackson A, Humbert MV, Pandey A, Bratcher H, Christodoulides M. 2015. Draft genome sequence of *Dichelobacter nodosus* ATCC 25549, strain VPI 2340 [11342], a bacterium causing footrot in sheep. Genome Announc 3(5):e01002-15. doi:10.1128/genomeA.01002-15.

Copyright © 2015 Jackson et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 3.0 Unported license.

Address correspondence to Myron Christodoulides, mc4@soton.ac.uk.

Dichelobacter nodosus, a Gram-negative, non-spore-forming obligate anaerobe, is the primary causative organism of footrot in ungulates (mammals with hooves, including cattle, sheep, deer, and goats) (1). The most well-known disease caused by *D. nodosus* is ovine footrot, a highly contagious necrotic disease of sheep hooves (2). A secondary role is postulated for *Fusobacterium necrophorum* through synergistic interactions with *D. nodosus* in footrot lesions (3). Knowledge of the genetic diversity and epidemiology of *D. nodosus* in ungulates is needed, given the significant animal welfare implications and economic costs associated with the disease. However, there is a significant lack of publically available complete genome sequences for *D. nodosus*.

D. nodosus (Beveridge, 1941) ATCC 25549, strain VPI 2340 [11342] (4) was grown for 7 to 10 days on Eugon Agar with defibrinated sheep blood (5% vol/vol) at 37°C in an anaerobic gas jar with the Anaerogen gas-generating system (Oxoid, United Kingdom). Genomic DNA was extracted using a standard phenolchloroform method, suspended in sterile ultrahigh-quality water (20 ng/ μ l), and purity confirmed by agarose (0.7% wt/vol) gel electrophoresis. DNA was sequenced using an Illumina HiSeq 2500 machine (Oxford Genomics Centre, Wellcome Trust Centre for Human Genetics, University of Oxford). The 150-bp shortread paired-end data were assembled with the de novo assembly algorithm Velvet (5) (version 1.2.08) combined with the VelvetOptimiser script (version 2.2.4). The minimum output contig size was set to 200 bp with the scaffolding option switched off; all other program settings were left at default. No read trimming was performed. The draft genome and annotations are available on the rMLST genome database (identification [ID] no. 121826) (http: //pubmlst.org/rmlst/) (6), which runs on the Bacterial Isolate Genome Sequence Database (BIGSdb) platform (7). Comparative genome analysis was run with the Genome Comparator tool implemented within BIGSdb using the gene-by-gene analysis approach (8).

The draft *D. nodosus* (Beveridge, 1941) ATCC 25549 genome has 36 contigs in its assembly, and the total length is 1,452,418 bp, coding for 1,271 nonredundant genes (with an additional 7 in-

complete). This draft genome sequence was compared to the genome sequence available for D. nodosus strain VCS1703A (9), of total length 1.39 Mb, encoding 1,276 genes (http://www.ncbi.nlm .nih.gov/nuccore/NC_009446.1). There are 1,263 identical genes (~98%) in both strains, but 13 genes in D. nodosus ATCC 25549encoding virulence-associated protein (Vap)F, VapA', VapI, RTX family protein, plasmid maintenance system killer family protein, an uncharacterized fimbrial protein, an uncharacterized membrane protein, and six hypothetical proteins-are absent. A comparative genome sequence study of 103 D. nodosus ovine isolates also showed high conservation (>95% sequence similarity) (10), but none of these sequences are available publicly. The D. nodosus ATCC 25549 genome contains 333 duplicated genes, and gene duplication is not observed in D. nodosus VCS1703A. Duplicated genes encode proteins associated with metabolism, transport, and efflux processes, outer membrane, and virulence, including porin(s), OmpA, proteases, VapB, VapG1, VapG2, VapG3, type IV fimbrial tip adhesin, and PilT. Increasing the number of available D. nodosus genome sequences, as well as the F. necrophorum coisolates, would provide insight into different virulence attributes and host-pathogen interactions and aid in the development of new vaccines.

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession no. LGVW00000000. The version described in this paper is version LGVW01000000.

ACKNOWLEDGMENTS

No funder was used to support this work. We thank Keith Jolley, University of Oxford, for providing access to the rMLST genome database for ID no. 121826 (http://pubmlst.org/rmlst/). We disclose no conflicts of interest.

REFERENCES

 Buller NB, Ashley P, Palmer M, Pitman D, Richards RB, Hampson DJ. 2010. Understanding the molecular epidemiology of the foot rot pathogen *Dichelobacter nodosus* to support control and eradication programs. J Clin Microbiol 48:877–882. http://dx.doi.org/10.1128/JCM.01355-09.

- Liu D, Yong WK. 1995. Molecular basis for the virulence of *Dichelobacter nodosus*. Trends Microbiol 3:474–475. http://dx.doi.org/10.1016/S0966 -842X(00)89013-7.
- 3. Bennett G, Hickford J, Sedcole R, Zhou H. 2009. *Dichelobacter nodosus*, *Fusobacterium necrophorum* and the epidemiology of footrot. Anaerobe 15:173–176. http://dx.doi.org/10.1016/j.anaerobe.2009.02.002.
- 4. Dewhirst FE, Paster BJ, La Fontaine S, Rood JI. 1990. Transfer of *Kingella indologenes* (Snell and Lapage 1976) to the genus *Suttonella* gen. nov. as *Suttonella indologenes* comb. nov.; transfer of *Bacteroides nodosus* (Beveridge 1941) to the genus *Dichelobacter* gen. nov. as *Dichelobacter nodosus* comb. nov.; and assignment of the genera *Cardiobacterium*, *Dichelobacter*, and *Suttonella* to *Cardiobacteriaceae* fam. nov. in the gamma division of Proteobacteria on the basis of 16S rRNA sequence comparisons. Int J Syst Bacteriol 40:426–433. http://dx.doi.org/10.1099/00207713 -40-4-426.
- Zerbino DR, Birney E. 2008. Velvet: algorithms for *de novo* short read assembly using de Bruijn graphs. Genome Res 18:821–829. http:// dx.doi.org/10.1101/gr.074492.107.
- Jolley KA, Bliss CM, Bennett JS, Bratcher HB, Brehony C, Colles FM, Wimalarathna H, Harrison OB, Sheppard SK, Cody AJ, Maiden MC. 2012. Ribosomal multilocus sequence typing: universal characterization

of bacteria from domain to strain. Microbiology 158:1005–1015. http://dx.doi.org/10.1099/mic.0.055459-0.

- Jolley KA, Maiden MC. 2010. BIGSdb: scalable analysis of bacterial genome variation at the population level. BMC Bioinformatics 11:595. http://dx.doi.org/10.1186/1471-2105-11-595.
- Jolley KA, Maiden MC. 2013. Automated extraction of typing information for bacterial pathogens from whole genome sequence data: *Neisseria meningitidis* as an exemplar. Euro Surveill 18:20379.
- Myers GS, Parker D, Al-Hasani K, Kennan RM, Seemann T, Ren Q, Badger JH, Selengut JD, Deboy RT, Tettelin H, Boyce JD, McCarl VP, Han X, Nelson WC, Madupu R, Mohamoud Y, Holley T, Fedorova N, Khouri H, Bottomley SP, Whittington RJ, Adler B, Songer JG, Rood JI, Paulsen IT. 2007. Genome sequence and identification of candidate vaccine antigens from the animal pathogen *Dichelobacter nodosus*. Nat Biotechnol 25:569–575. http://dx.doi.org/10.1038/nbt1302.
- Kennan RM, Gilhuus M, Frosth S, Seemann T, Dhungyel OP, Whittington RJ, Boyce JD, Powell DR, Aspán A, Jørgensen HJ, Bulach DM, Rood JI. 2014. Genomic evidence for a globally distributed, bimodal population in the ovine foot rot pathogen *Dichelobacter nodosus*. mBio 5:e01821-14. http://dx.doi.org/10.1128/mBio.01821-14.