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Draft Genome Sequence of *Enterococcus faecium* PC4.1, a Clade B Strain Isolated from Human Feces

Páraic Ó Cuív,[a,b*] Eline S. Klaassens,[a,b*] Wendy J. Smith,[a,b] Stanislas Mondot,[a,b*] A. Scott Durkin,[c] Derek M. Harkins,[c] Les Foster,[c*] Jamison McCorrison,[c] Manolito Torralba,[c] Karen E. Nelson,[c] Mark Morrison[a,b,d*]

CSIRO Preventative Health Flagship Research Program, Queensland, Australia[a]; CSIRO Animal, Food and Health Sciences, St. Lucia, Queensland, Australia[b]; J. Craig Venter Institute, Rockville, Maryland, USA[c]; The Ohio State University, Columbus, Ohio, USA[d]

* Present address: Eline S. Klaassens, School of Molecular Bioscience, University of Sydney, Sydney, Australia; Stanislas Mondot, Institut Curie, Paris, France; Les Foster, Howard Hughes Medical Institute, Janelia Farm Research Campus, Ashburn, Virginia, USA; Páraic Ó Cuív and Mark Morrison, The University of Queensland, Diamantina Institute, Queensland, Australia.

***Enterococcus faecium* is commonly isolated from the human gastrointestinal tract; however, important intraspecies variations exist with relevance for host health and well-being. Here, we describe the draft genome sequence of *E. faecium* PC4.1, a clade B strain isolated from human feces.**

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Address correspondence to Páraic Ó Cuív, p.ocuiv@uq.edu.au.

The facultative anaerobic bacterium *Enterococcus faecium* is among the early colonizers of the infant gut, where it has been shown to be transferred via the mother's breast milk (1, 2). *E. faecium* is also one of the most abundant enterococcal species in the adult colon, where it likely plays an important role in maintaining host health and well-being, as has been suggested by human and animal studies (3–5). Significant intraspecies variations are known to exist; however, despite having an open pangenome (6), a recent comparative genomic analysis of *E. faecium* revealed the existence of only two distinct phylogenetic clades, termed clades A and B (7). Interestingly, most of the clinical and "healthy" isolates can be assigned to clade A and clade B, respectively, with clade B strains serving as important donors of DNA for clade A strains (8). Here, we describe the draft genome sequence of *E. faecium* PC4.1, isolated as part of the Australian Human Gut Microbiome Project, in order to provide new insights into the functional versatility of clade B strains.

E. faecium PC4.1 was isolated from a pooled fecal sample collected from healthy human subjects by plating aerobically on bile esculin azide agar. *E. faecium* isolates were distinguished from *Enterococcus faecalis* isolates by their β-galactosidase activity, as assessed on brain heart infusion agar supplemented with 5-bromo-4-chloro-3-indolyl-β-D-galactopyranoside. A 454 Life Sciences GS FLX system was used at the J. Craig Venter Institute (JCVI) to generate 2,811,160 bp of shotgun genomic DNA sequence at 20.7× coverage. Next, the Newbler Assembler version 1.1 was used to assemble the individual sequence reads, generating 78 contigs, with a contig N_{50} of 100.5 kb and with the largest contig assembled of approximately 272.2 kb. Finally, the JCVI prokaryotic annotation pipeline was used to annotate the DNA sequences.

The draft genome has a G+C content of 37.98% and contains 2,739 genes, including 2,695 protein-coding genes and 44 structural RNAs. As expected, several niche factors (9) were identified that likely contribute to its effective colonization and persistence in the human gut, including protein orthologs with predicted roles in binding host structural factors, including collagen, fibronectin, and nidogen. In addition, we also identified a putative lectin, which is consistent with the ability of enterococci to bind host sugar moieties (10). *E. faecium* PC4.1 encodes a candidate autolysin necessary for DNA release and biofilm formation (11); however, it does not encode the phosphotransferase system that is enriched in clinical isolates (7), nor does it encode the gelatinase and serine proteinase typically found in virulent enterococcal strains.

The draft *E. faecium* PC4.1 genome has provided an insight into the factors that support the ability of clade B isolates to colonize and persist in the human gut. Future studies should determine the genome phylogeny of putative pathogenic strains, including Crohn's disease-associated strains (12, 13), and compare their functional potentials to those of healthy clade B isolates. The genome sequence of *E. faecium* PC4.1 and those of other healthy gut isolates will serve as valuable resources in this respect.

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession no. ADMM00000000. The version described here is the first version, ADMM01000000.

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REFERENCES

1. **Martín R, Langa S, Reviriego C, Jiménez E, Marín ML, Xaus J, Fernández L, Rodríguez JM.** 2003. Human milk is a source of lactic acid bacteria for the infant gut. J. Pediatr. **143:**754–758. http://dx.doi.org/10.1016/j.jpeds.2003.09.028.
2. **Albesharat R, Ehrmann MA, Korakli M, Yazaji S, Vogel RF.** 2011. Phenotypic and genotypic analyses of lactic acid bacteria in local fermented food, breast milk and faeces of mothers and their babies. Syst. Appl. Microbiol. **34:**148–155. http://dx.doi.org/10.1016/j.syapm.2010.12.001.
3. **Jin LZ, Marquardt RR, Zhao X.** 2000. A strain of *Enterococcus faecium* (18C23) inhibits adhesion of enterotoxigenic *Escherichia coli* K88 to porcine small intestine mucus. Appl. Environ. Microbiol. **66:**4200–4204. http://dx.doi.org/10.1128/AEM.66.10.4200-4204.2000.
4. **Benyacoub J, Czarnecki-Maulden GL, Cavadini C, Sauthier T, Anderson RE, Schiffrin EJ, von der Weid T.** 2003. Supplementation of food with *Enterococcus faecium* (SF68) stimulates immune functions in young dogs. J. Nutr. **133:**1158–1162.
5. **Wunderlich PF, Braun L, Fumagalli I, D'Apuzzo V, Heim F, Karly M, Lodi R, Politta G, Vonbank F, Zeltner L.** 1989. Double-blind report on the efficacy of lactic acid-producing *Enterococcus* SF68 in the prevention of antibiotic-associated diarrhoea and in the treatment of acute diarrhoea. J. Int. Med. Res. **17:**333–338.
6. **van Schaik W, Top J, Riley DR, Boekhorst J, Vrijenhoek JE, Schapendonk CM, Hendrickx AP, Nijman IJ, Bonten MJ, Tettelin H, Willems RJ.** 2010. Pyrosequencing-based comparative genome analysis of the nosocomial pathogen *Enterococcus faecium* and identification of a large transferable pathogenicity island. BMC Genomics **11:**239. http://dx.doi.org/10.1186/1471-2164-11-239.
7. **Zhang X, Top J, de Been M, Bierschenk D, Rogers M, Leendertse M, Bonten MJ, van der Poll T, Willems RJ, van Schaik W.** 2013. Identification of a genetic determinant in clinical *Enterococcus faecium* strains that contributes to intestinal colonization during antibiotic treatment. J. Infect. Dis. **207:**1780–1786. http://dx.doi.org/10.1093/infdis/jit076.
8. **de Been M, van Schaik W, Cheng L, Corander J, Willems RJ.** 2013. Recent recombination events in the core genome are associated with adaptive evolution in *Enterococcus faecium*. Genome Biol Evol. **5:**1524–1535. http://dx.doi.org/10.1093/gbe/evt111.
9. **Hill C.** 2012. Virulence or niche factors: what's in a name? J. Bacteriol. **194:**5725–5727. http://dx.doi.org/10.1128/JB.00980-12.
10. **Štyriak I, Lauková A, Ljungh Å.** 2002. Lectin-like binding and antibiotic sensitivity of enterococci from wild herbivores. Microbiol. Res. **157:**293–303. http://dx.doi.org/10.1078/0944-5013-00166.
11. **Paganelli FL, Willems RJ, Jansen P, Hendrickx A, Zhang X, Bonten MJ, Leavis HL.** 2013. *Enterococcus faecium* biofilm formation: identification of major autolysin $AtlA_{Efm}$, associated Acm surface localization, and $AtlA_{Efm}$-independent extracellular DNA release. mBio **4:**e00154-13. http://dx.doi.org/10.1128/mBio.00154-13.
12. **Kang S, Denman SE, Morrison M, Yu Z, Dore J, Leclerc M, McSweeney CS.** 2010. Dysbiosis of fecal microbiota in Crohn's disease patients as revealed by a custom phylogenetic microarray. Inflamm. Bowel Dis. **16:**2034–2042. http://dx.doi.org/10.1002/ibd.21319.
13. **Mondot S, Kang S, Furet JP, Aguirre de Carcer D, McSweeney C, Morrison M, Marteau P, Doré J, Leclerc M.** 2011. Highlighting new phylogenetic specificities of Crohn's disease microbiota. Inflamm. Bowel Dis. **17:**185–192. http://dx.doi.org/10.1002/ibd.21436.