





# Complete Genome Sequence of *Enterococcus faecalis* Strain SGAir0397, Isolated from a Tropical Air Sample Collected in Singapore

 Rikky W. Purbojati,<sup>a</sup> Daniela I. Drautz-Moses,<sup>a</sup> Akira Uchida,<sup>a</sup> Anthony Wong,<sup>a</sup> Megan E. Clare,<sup>a</sup> Kavita K. Kushwaha,<sup>a</sup> Alexander Putra,<sup>a</sup> Balakrishnan N. V. Premkrishnan,<sup>a</sup> Cassie E. Heinle,<sup>a</sup> Ngu War Aung,<sup>a</sup> Vineeth Kodengil Vettath,<sup>a</sup>  
 Ana Carolina M. Junqueira,<sup>b</sup> Stephan C. Schuster<sup>a</sup>

<sup>a</sup>Singapore Centre for Environmental Life Sciences Engineering, Nanyang Technological University, Singapore

<sup>b</sup>Departamento de Genética, Instituto de Biologia, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil

**ABSTRACT** *Enterococcus faecalis* strain SGAir0397 was isolated from a tropical air sample collected in Singapore. Its genome was assembled using single-molecule real-time sequencing data and comprises one circular chromosome with a length of 2.69 Mbp. The genome contains 2,595 protein-coding genes, 59 tRNAs, and 12 rRNAs.

*Enterococcus faecalis*, a Gram-positive, coccus-shaped, facultative anaerobe, belongs to the phylum *Firmicutes* and is commonly found in the gastrointestinal tract of humans (1) and animals (2). It is typically found as a commensal in the human gut but has the potential to cause nosocomial infections (3) as a response to environmental changes, such as antibiotic-mediated alteration of the intestinal microbiome (4) or migration to the bloodstream or other sites in the human body (5).

One common mechanism of dispersal is through the release of human and animal fecal matter from the gastrointestinal tract (6). *Enterococcus faecalis* has been identified and isolated from exogenous environments such as plants and soils (7), marine water and sediment (8), freshwater (9, 10), and sewage (11). Due to its close association with the gastrointestinal tract, traces of this bacterium in surface water are commonly used as an indicator of human/animal fecal contamination (10, 12).

The strain SGAir0397 was isolated from an air sample collected in Singapore (global positioning system [GPS] location 1.344°N, 103.68°E) using an Andersen single-stage impactor (SKC, USA). The collected air was impacted onto Marine agar (Becton, Dickinson, USA), and further isolation was carried out by cultivation on tryptic soy agar (Becton, Dickinson) at 30°C until axenic growing colonies were obtained. A single colony was inoculated in lysogeny broth (Becton, Dickinson) and grown overnight for genomic DNA extraction. Genomic DNA was purified using the Wizard genomic DNA purification kit (Promega, USA), according to the manufacturer's protocol. Library preparation was performed with the SMRTbell template prep kit 1.0 (Pacific Biosciences, USA), followed by single-molecule real-time (SMRT) sequencing on the Pacific Biosciences RS II platform.

A total of 80,144 subreads were used for *de novo* assembly with Hierarchical Genome Assembly Process (HGAP) version 3 (13) and polished using Quiver (13) within the PacBio SMRT Analysis 2.3.0 assembly protocol. The polished assembly was then circularized and reoriented with Circlator 1.1.4 (14), resulting in one circular chromosome contig with a length of 2,696,714 bp (217-fold coverage). The chromosomal contig showed a mean G+C content of 37.8%.

Taxonomic identification was performed using the full-length 16S rRNA gene sequence, predicted with Barrnap 0.7 (15), and by determining the average nucleotide identity (ANI) of the genome. The BLASTn (16) result showed 100% sequence identity

**Citation** Purbojati RW, Drautz-Moses DI, Uchida A, Wong A, Clare ME, Kushwaha KK, Putra A, Premkrishnan BNV, Heinle CE, Aung NW, Vettath VK, Junqueira ACM, Schuster SC. 2019. Complete genome sequence of *Enterococcus faecalis* strain SGAir0397, isolated from a tropical air sample collected in Singapore. *Microbiol Resour Announc* 8:e00593-19. <https://doi.org/10.1128/MRA.00593-19>.

**Editor** David Rasko, University of Maryland School of Medicine

**Copyright** © 2019 Purbojati et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Stephan C. Schuster, [SCSchuster@ntu.edu.sg](mailto:SCSchuster@ntu.edu.sg).

**Received** 29 May 2019

**Accepted** 22 July 2019

**Published** 15 August 2019

to *Enterococcus faecalis* strain CVM N48037F. Meanwhile, ANI analysis (17) resulted in a closest similarity of 98.9% to the *Enterococcus faecalis* ATCC 19433 genome.

The genome was annotated with the RASTtk (18) Web server. A total of 2,595 coding sequences (CDS), 12 rRNA operons (5S, 16S, and 23S rRNAs), and 59 tRNAs were predicted. A CRISPR array with the repeat element GTTTTGGTACCATTCTAAACAACATG ACTCTAAAAC spans a total of 432 bp in the chromosome. antiSMASH 5.0.0rc1 analysis (19) resulted in the prediction of a secondary metabolite biosynthesis prediction gene cluster of bacteriocin. Meanwhile, antibiotic resistance gene analysis with ResFinder 2.1 (20) predicted a gene associated with resistance to macrolide, lincosamide, and streptogramin B antibiotics. Default parameters were used for all software programs, unless otherwise specified.

The isolation of this *Enterococcus faecalis* strain SGAir0397 from the air may provide additional insight into the mode of dissemination and transfer between reservoirs, which could be clinically relevant in the context of hospital-acquired enterococcal infections.

**Data availability.** The complete genome sequence of *Enterococcus faecalis* strain SGAir0397 has been deposited in the NCBI under accession number [CP039434](https://doi.org/10.1093/nar/gkz310), and the raw sequencing data have been deposited in the NCBI SRA under accession number [SRR9043821](https://doi.org/10.1093/nar/gkz310).

## ACKNOWLEDGMENTS

This work was supported by a Singapore Ministry of Education Academic Research Fund tier 3 grant (MOE2013-T3-1-013).

We thank Anjali Bansal Gupta for providing her input and for proofreading the manuscript.

## REFERENCES

- Tannock GW, Cook G. 2002. Enterococci as members of the intestinal microflora of humans, p 101–132. *In* Gilmore MS, Clewell DB, Courvalin P, Dunny GM, Murray BE, Rice LB (ed), *The enterococci: pathogenesis, molecular biology, and antibiotic resistance*. ASM Press, Washington, DC.
- Mundt JO. 1963. Occurrence of enterococci in animals in a wild environment. *Appl Microbiol* 11:136–140.
- Gilmore MS, Lebreton F, van Schaik W. 2013. Genomic transition of enterococci from gut commensals to leading causes of multidrug-resistant hospital infection in the antibiotic era. *Curr Opin Microbiol* 16:10–16. <https://doi.org/10.1016/j.mib.2013.01.006>.
- Ubeda C, Taur Y, Jenq RR, Equinda MJ, Son T, Samstein M, Viale A, Socci ND, van den Brink MR, Kamboj M, Pamer EG. 2010. Vancomycin-resistant *Enterococcus* domination of intestinal microbiota is enabled by antibiotic treatment in mice and precedes bloodstream invasion in humans. *J Clin Invest* 120:4332–4341. <https://doi.org/10.1172/JCI43918>.
- Arias CA, Murray BE. 2012. The rise of the *Enterococcus*: beyond vancomycin resistance. *Nat Rev Microbiol* 10:266–278. <https://doi.org/10.1038/nrmicro2761>.
- Byappanahalli MN, Nevers MB, Korajkic A, Staley ZR, Harwood VJ. 2012. Enterococci in the environment. *Microbiol Mol Biol Rev* 76:685–706. <https://doi.org/10.1128/MMBR.00023-12>.
- Mundt JO, Coggin JH, Jr, Johnson LF. 1962. Growth of *Streptococcus faecalis* var. *liquefaciens* on plants. *Appl Microbiol* 10:552–555.
- Ferguson D, Moore D, Getrich M, Zhouwandai M. 2005. Enumeration and speciation of enterococci found in marine and intertidal sediments and coastal water in southern California. *J Appl Microbiol* 99:598–608. <https://doi.org/10.1111/j.1365-2672.2005.02660.x>.
- Gary HL, Adams JC. 1985. Indicator bacteria in water and stream sediments near the snowy range in southern Wyoming. *Water Air Soil Pollut* 25:133–144. <https://doi.org/10.1007/BF00568382>.
- Anderson KL, Whitlock JE, Harwood VJ. 2005. Persistence and differential survival of fecal indicator bacteria in subtropical waters and sediments. *Appl Environ Microbiol* 71:3041–3048. <https://doi.org/10.1128/AEM.71.6.3041-3048.2005>.
- Berg G, Dahling DR, Brown GA, Berman D. 1978. Validity of fecal coliforms, total coliforms, and fecal streptococci as indicators of viruses in chlorinated primary sewage effluents. *Appl Environ Microbiol* 36:880–884.
- Boehm AB, Sassoubre LM. 2014. Enterococci as indicators of environmental fecal contamination. *In* Gilmore MS, Clewell DB, Ike Y (ed), *Enterococci: from commensals to leading causes of drug resistant infection*. Massachusetts Eye and Ear Infirmary, Boston, MA.
- Chin CS, Alexander DH, Marks P, Klammer AA, Drake J, Heiner C, Clum A, Copeland A, Huddleston J, Eichler EE, Turner SW, Korlach J. 2013. Nonhybrid, finished microbial genome assemblies from long-read SMRT sequencing data. *Nat Methods* 10:563. <https://doi.org/10.1038/nmeth.2474>.
- Hunt M, Silva ND, Otto TD, Parkhill J, Keane JA, Harris SR. 2015. Circlator: automated circularization of genome assemblies using long sequencing reads. *Genome Biol* 16:294. <https://doi.org/10.1186/s13059-015-0849-0>.
- Seemann T. 2014. Prokka: rapid prokaryotic genome annotation. *Bioinformatics* 30:2068–2069. <https://doi.org/10.1093/bioinformatics/btu153>.
- Altschul SF, Madden TL, Schaffer AA, Zhang J, Zhang Z, Miller W, Lipman DJ. 1997. Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. *Nucleic Acids Res* 25:3389–3402. <https://doi.org/10.1093/nar/25.17.3389>.
- Shen L, Liu Y, Xu B, Wang N, Zhao H, Liu X, Liu F. 2017. Comparative genomic analysis reveals the environmental impacts on two *Arcticibacter* strains including sixteen *Sphingobacteriaceae* species. *Sci Rep* 7:2055. <https://doi.org/10.1038/s41598-017-02191-4>.
- Brettin T, Davis JJ, Disz T, Edwards RA, Gerdes S, Olsen GJ, Olson R, Overbeek R, Parrello B, Pusch GD, Shukla M, Thomson JA, Stevens R, Vonstein V, Wattam AR, Xia F. 2015. RASTtk: a modular and extensible implementation of the RAST algorithm for building custom annotation pipelines and annotating batches of genomes. *Sci Rep* 5:8365. <https://doi.org/10.1038/srep08365>.
- Blin K, Shaw S, Steinke K, Villebro R, Ziemert N, Lee SY, Medema MH, Weber T. 2019. antiSMASH 5.0: updates to the secondary metabolite genome mining pipeline. *Nucleic Acids Res* 47:W81–W87. <https://doi.org/10.1093/nar/gkz310>.
- Zankari E, Hasman H, Cosentino S, Vestergaard M, Rasmussen S, Lund O, Aarestrup FM, Larsen MV. 2012. Identification of acquired antimicrobial resistance genes. *J Antimicrob Chemother* 67:2640–2644. <https://doi.org/10.1093/jac/dks261>.