Draft Genome Sequences of Six Enterococcus faecalis Strains Isolated from Malaysian Clinical and Environmental Origins

Diane Sunira Daniel,a,b Han Ming Gan,a,b Sui Mae Lee,a,b,c Gary A. Dykes,c Sadequr Rahman,a,b
School of Science, Monash University, Bandar Sunway, Malaysia; Tropical Medicine and Biology Platform, Monash University, Bandar Sunway, Malaysia; School of Public Health, Curtin University, Perth, Western Australia

ABSTRACT Enterococcus faecalis is known to cause a variety of nosocomial infections, including urinary tract infections. Antibiotic resistance and virulence properties in this species are of public concern. The draft genome sequences of six E. faecalis strains isolated from clinical and environmental sources in Malaysia are presented here.

Enterococcus faecalis is an opportunistic pathogen that is often recovered from urinary tract infections (UTIs). E. faecalis is known to cause infections mainly due to the expression of virulence factors associated with adherence of mucosal and abiotic surfaces (1). The number of complete or draft genome sequences available for E. faecalis as of April 2017 is 503, comprising the bulk of enterococcal genome sequences available. However, there is a poor representation of genomic sequences for enterococci from Malaysia with only approximately seven assemblies reported (2). In this study, six E. faecalis strains, designated S12, S13, S14, S15, S16, and S17, were sequenced. These six strains were previously isolated from water sources, farm animal feces, and UTI patients in Malaysia and selected on the basis of different pulsed-field gel electrophoresis pulsotypes reported in a previously published experiment (3), and different biofilm and attachment properties (our unpublished data).

Genomic DNA was extracted using a GF-1 bacterial DNA extraction kit (Vivantis, Malaysia), tagmented with Nextera XT (Illumina, USA) according to the manufacturer’s instructions and sequenced on the MiSeq desktop sequencer located at the Monash University Malaysia Genomics Facility (2 × 250-bp run configuration). The raw reads generated were trimmed (quality score limit of 0.05) and assembled de novo using CLC Genomics Workbench version 7.0 (CLC bio, Denmark). Genome annotation was performed using the NCBI Prokaryotic Genome Annotation Pipeline and the Rapid Annotations using Subsystems Technology server (4, 5). The identification of acquired antibiotic resistance and virulence genes was performed with the web tools ResFinder and VirulenceFinder, respectively (6, 7). Contigs coding for each antibiotic resistance and virulence gene (represented by contig accession number) were filtered based on 90% identity to the reference sequence and are presented in Table 1.

Genomic statistics—mean coverage, \( N_{50} \) contig length, number of contigs, assembly size, number of coding sequences, and number of tRNAs and rRNAs for the six assembled genomes are provided in Table 1. An orthologous average nucleotide identity tool based on OrthoANI values revealed a >98% similarity of all six strains to the whole genome of E. faecalis ATCC 19433 (PRJNA157741) (8).

Strain S14 harbors the complete gene cassette for vancomycin resistance, corroborating a previous wet lab report that recorded a MIC value of 64 \( \mu g/mL \) (3), whereas
strains S12, S13, S14, S15, S16, and S17 have been deposited in DDBJ/ENA/GenBank under the accession numbers listed in Table 1.

ACKNOWLEDGMENTS

The project was supported by the School of Science, Monash University Malaysia. We are grateful to Yin Peng Lee from the Monash University Malaysia Tropical Medicine and Biology Multidisciplinary Platform for her assistance in next-generation sequencing.

REFERENCES

1. Maryam D, Ozlem GE, M NA, Fatma NY, Evrim GA, Nefise A. 2014. The interactions between esp, fsr, gelE genes and biofilm formation and pIge analysis of clinical Enterococcus faecium strains. Afr J Microbiol Res 8:129–137. https://doi.org/10.5897/AJMR2013.6257.

2. Daniel DS, Lee SM, Gan HM, Dykes GA, Rahman S. 2017. The public health risks of multiple-drug resistant (MDR) Enterococcus spp. in Southeast Asia. Appl Environ Microbiol 81:6090 – 6097. https://doi.org/10.1128/AEM.01741-15.

3. Daniel DS, Lee SM, Gan HM, Dykes GA, Rahman S. 2017. Genetic diversity of Enterococcus faecalis isolated from environmental, animal and clinical sources in Malaysia. J Infect Public Health [Epub ahead of print.] https://doi.org/10.1016/j.jiph.2017.02.006.

4. Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeek RA, McNeil LK, Paarmann D, Paczian T, Parrello B, Pusch GD, Reich C, Stevens R, Yooseph S, Vassieva O, White O, Wilson SH, Zhuang Z, Zaslawsky L, Lomsadze A, Pruitt KD, Borodovsky M, Nisbet E, et al. 2008. The RAST server: rapid annotations using subsystems technology. BMC Genomics 9:75. https://doi.org/10.1186/1471-2164-9-75.

5. Tatusova T, DiCuccio M, Badretdin A, Chetvernin V, Nawrocki EP, Zaslavsky L, Lomsadze A, Pruitt KD, Borodovsky M, Ostell J. 2016. NCBI prokaryotic genome annotation pipeline. Nucleic Acids Res 44: 6614–6624. https://doi.org/10.1093/nar/gkw569.

6. Zankari E, Hasman H, Cosentino S, Vestergaard M, Rasmussen S, Lund O, Aarestrup FM. 2012. Identification of acquired antimicrobial resistance genes. J Antimicrob Chemother 67:2640 –2644. https://doi.org/10.1093/jac/dks261.

7. Joensen KG, Scheutz F, Lund O, Hasman H, Kaas RS, Nielsen EM, Aarestrup FM. 2014. Real-time whole genome sequencing for routine typing, surveillance, and outbreak detection of verotoxigenic Escherichia coli. J Clin Microbiol 52:1501–1510. https://doi.org/10.1128/JCM.03617-13.

8. Lee I, Kim YO, Park SC, Chun J. 2015. OrthoANI: an improved algorithm and software for calculating average nucleotide identity. Int J Syst Evol Microbiol [Epub ahead of print.] https://doi.org/10.1099/ijsem.0.000760.

9. Anderson AC, Jonas D, Huber I, Kargianni L, Wölfer J, Herrmann A, Arweiler N, Vach K, Wittmer A, Al-Abdul Aziz. 2015. Enterococcus faecalis from Food, clinical specimens and oral sites: prevalence of virulence factors in association with biofilm formation. Front Microbiol 6:1534. https://doi.org/10.3389/fmicb.2015.01534.