

Draft Genome Sequence of the Quality Control Strain *Enterococcus faecalis* ATCC 29212

Eun Bae Kim, Lauren M. Kopit, Linda J. Harris, and Maria L. Marco

Department of Food Science and Technology, University of California, Davis, California, USA

***Enterococcus faecalis* ATCC 29212, a vancomycin-sensitive strain, has been extensively used as a representative control strain for clinical and laboratory experiments. Here we report the draft genome and annotation of this strain, containing 3,027,060 bp, with a G+C content of 37.2% in 126 contigs (≥ 500 bp).**

Enterococcus faecalis is a Gram-positive lactic acid bacterium found in animal intestines and a variety of food- and plant-associated environments and is increasingly recognized as a nosocomial pathogen. *E. faecalis* ATCC 29212, an isolate from human urine (1), is used as a control strain to assess the quality of commercially prepared microbiological culture media (9), susceptibility testing of pathogens to antibiotics (12, 13), commercialized biochemical identification of bacterial species (API, Vitek, etc.), and microbiological quality testing of food and animal feeding stuffs (7). The heat tolerance of this strain is similar to that of *Salmonella enterica* serovar Enteritidis on inoculated almonds in hot water treatments (6), indicating that this organism might be used as a surrogate in challenge tests evaluating heat-processing protocols designed to kill food-borne pathogens. Contrary to such broad general uses of *E. faecalis* ATCC 29212 as a standard control in clinical and food safety tests, basic information regarding the genetic composition of this strain is lacking. The need for genome-level information is indicated by the fact that *E. faecalis* strains are now regarded as opportunistic pathogens and that hospital-associated strains typically share a similar genetic composition and resistance to clinically relevant antibiotics (2). Here, we sequenced and annotated the genome of ATCC 29212 to enable broader applications of this organism for comparison of genotypic differences among members of this species.

We received *E. faecalis* ATCC 29212 from the American Type Culture Collection (ATCC). The strain was grown overnight in brain heart infusion (BHI) medium, and genomic DNA was extracted using the DNeasy blood and tissue kit (Qiagen). The genomic DNA was sequenced using the Ion Torrent PGM sequencer (200-bp library) at EdgeBio (Gaithersburg, MD) (10). The sequences were quality filtered, resulting in 317 Mbp in 1,558,529 reads, representing a 104-fold coverage of the genome. The reads were then assembled by Ray 1.7 (4), with a k-mer size of 31 bp to generate 126 contigs (≥ 500 bp; total length, 3,027,060 bp; N_{50} length, 52,252 bp; average length, 24,024 bp; G+C content, 37.2%). A total of 2,984 coding sequences (CDS) and 50 structural RNAs (46 tRNAs) were predicted by rapid annotation using subsystem technology (RAST) (3). Among the CDS, 1,339 CDS (45% of total) were distributed over 343 functional gene categories in RAST, which is similar to the result for other *E. faecalis* genomes (data not shown).

Multilocus sequence typing (MLST) based on seven house-keeping genes (11) showed that ATCC 29212 belongs to sequence type 30 (ST30). ST30 contains fewer clinical isolates than other *E. faecalis* STs in the MLST database ($P = 0.00427$) (11). Despite its

association with the less virulent group, this strain contains 20 out of 36 genetic loci associated with *E. faecalis* virulence (2), including endocarditis-associated collagen adhesion (*ace*) and hemolysin-cytolysin (*cyl* operon). Genes coding for tetracycline resistance (*tetM*) were found, but evidence for vancomycin resistance was lacking, and these findings are in agreement with the levels of antibiotic resistance known for this strain (5, 8, 12, 13). The genome sequence of *E. faecalis* ATCC 29212 provides a map for the reevaluation of the phenotypes that have been reported for this strain and for the assessment of pathogenicity.

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession number [ALOD000000000](http://www.ncbi.nlm.nih.gov/nuccore/ALOD000000000). The version described in this paper is the first version, ALOD010000000.

ACKNOWLEDGMENTS

This work was supported by a grant from the Almond Board of California.

We acknowledge Vanessa Morales for her assistance with the antibiotic resistance tests.

REFERENCES

- Andrewes FW, Horder TJ. 1906. A study of the streptococci pathogenic for man. *Lancet* 168:852–855.
- Arias CA, Murray BE. 2012. The rise of the *Enterococcus*: beyond vancomycin resistance. *Nat. Rev. Microbiol.* 10:266–278.
- Aziz R, et al. 2008. The RAST server: rapid annotations using subsystems technology. *BMC Genomics* 9:75.
- Boisvert S, Laviolette F, Corbeil J. 2010. Ray: simultaneous assembly of reads from a mix of high-throughput sequencing technologies. *J. Comput. Biol.* 17:1519–1533.
- Brown SD, Traczewski MM. 2007. Comparative *in vitro* antimicrobial activity of tigecycline, a new glycylcycline compound, in freshly prepared medium and quality control. *J. Clin. Microbiol.* 45:2173–2179.
- Du WX, Abd SJ, McCarthy KL, Harris LJ. 2010. Reduction of *Salmonella* on inoculated almonds exposed to hot oil. *J. Food Prot.* 73:1238–1246.
- International Organization for Standardization. 2003. Microbiology of food and animal feeding stuffs—guidelines on preparation and production of culture media. ISO, Geneva, Switzerland.
- Johnston LM, Jaykus L-A. 2004. Antimicrobial resistance of *Enterococcus* species isolated from produce. *Appl. Environ. Microbiol.* 70:3133–3137.
- Krisner K, et al. 2004. Quality control for commercially prepared micro-

Received 13 August 2012 Accepted 24 August 2012

Address correspondence to Maria L. Marco, mmarco@ucdavis.edu.

Copyright © 2012, American Society for Microbiology. All Rights Reserved.

doi:10.1128/JB.01423-12

- biological culture media; approved standard—third edition. Clinical and Laboratory Standards Institute, Wayne, PA.
10. **Rothberg JM, et al.** 2011. An integrated semiconductor device enabling non-optical genome sequencing. *Nature* 475:348–352.
 11. **Urwin R, Maiden MCJ.** 2003. Multi-locus sequence typing: a tool for global epidemiology. *Trends Microbiol.* 11:479–487.
 12. **Wikler MA, et al.** 2007. Performance standards for antimicrobial susceptibility testing; seventeenth informational supplement. Clinical and Laboratory Standards Institute, Wayne, PA.
 13. **Wikler MA, et al.** 2006. Performance standards for antimicrobial disk susceptibility tests; approved standard—ninth edition. Clinical and Laboratory Standards Institute, Wayne, PA.