Complete Genome Sequence of the Commensal Enterococcus faecalis 62, Isolated from a Healthy Norwegian Infant^{∇}

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The genome of Enterococcus faecalis 62, a commensal isolate from a healthy Norwegian infant, revealed multiple adaptive traits to the gastrointestinal tract (GIT) environment and the milk-containing diet of breast-fed infants. Adaptation to a commensal existence was emphasized by lactose and other carbohydrate metabolism genes within genomic islands, accompanied by the absence of virulence traits.

Enterococcus faecalis is a multifaceted lactic acid bacterium with an intimate relationship to human health and disease (7). E. faecalis is among the earliest bacteria that colonize the gastrointestinal tracts of newborns (1), and for most people, E. faecalis is part of the normal commensal intestinal microflora (14). Remarkably, certain strains are used as probiotics (5), but E. faecalis is also a prominent cause of multiresistant nosocomial infections (9, 18). This situation has spurred a tremendous interest in the composition of E. faecalis genomes, with the number of publicly available draft genomes exceeding 50 (2, 15; http://www.ncbi.nlm.nih.gov/genomepri/20875), but only the V583 and OG1RF genomes have been completely determined (16).

Here we report the complete genome sequence of the commensal E. faecalis 62, which was isolated from a healthy Norwegian infant (20). This isolate has the multilocus sequence type ST66, which is not part of any clonal complex related to nosocomial infections (20). Sequencing was performed at Eurofins MWG-Operon, Germany, using 454 Life Sciences pyrosequencing. Three data sets were obtained: (i) GS FLX, with 235,561 shotgun reads and an average read length of 230 nucleotides (nt); (ii) GS FLX Titanium, with 111,123 shotgun reads and an average read length of 390 nt; (iii) GS 20, with 113,078 reads from a 2-kb paired-end library (of which 56,539 were paired reads, equaling a 50-fold clone coverage) and an average read length of 99 nt (total, ~101 Mb; ~32-fold coverage). Contigs were generated and assembled into scaffolds by using the Newbler Assembler software (454 Life Sciences) via the Bioportal server (www.bioportal.uio.no).

The genome structure was compared to reference strain

V583 (16) by a progressiveMauve alignment (4). Physical gaps, repeats, and assembly ambiguities were corrected through sequencing of PCR products. Open reading frame (ORF) calling and annotation were first performed using the Institute for Genome Sciences (IGS) annotation service (University of Maryland). Manual annotation was performed using Pfam (6), InterProScan (17), and BLAST comparisons to completely sequenced genomes at NCBI (3).

The genome of E. faecalis 62 consists of a single, circular chromosome (2,988,673 bp; 37.2% GC content), a pseudotemperate linear bacteriophage (EF62phi), and three plasmids (EF62pA, EF62pB, and EF62pC). The chromosome contains 2,893 protein-encoding genes, 54 tRNA-encoding genes, and 4 rRNA-encoding operons. The existence of a linear pseudotemperate Podoviridae bacteriophage, EF62¢ (30,505 bp), is new to E. faecalis. This phage replicates extrachromosomally as a linear DNA molecule via a repB gene, while a toxin-antitoxin system probably ensures temperate state maintenance. The EF62pA plasmid (5,143 bp) is virtually identical to E. faecalis plasmid pS86 (10). EF62pB (51,104 bp) is a conjugative plasmid highly similar to pCF10 (8) but lacks the 18-kb Tn916 element. Notably, a Tn916 element is found in the chromosome of E. faecalis 62. EF62pC (55,393 bp) resembles pAM737-type plasmids associated with the pathogenicity island (PAI) element (11, 12, 19).

With respect to virulence traits, E. faecalis 62 contains the gelatinase gelE and serine proteinase sprE genes but displays a gelatinase-negative phenotype (24), caused by a deletion in the fsr regulatory system and consistent with previous observations (13). The genomic region corresponding to the enterococcus PAI in E. faecalis 62 lacks elements involved in virulence, including cytolysin, compared to V583 or MMH594 (19). This genomic island does, however, contain the enterococcal surface protein gene (esp), the bile salt hydrolase gene (cbh), and lactose metabolic pathway genes (lacABCDEFG).

These results illustrate the complexity involved in elucidation of the genomic differences that distinguish commensal and pathogenic E. faecalis isolates.

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