



Complete Genome Sequence of a Human Norovirus Strain from the United States Classified as Genotype GII.P6_GII.6

Haifeng Chen,^a Shiliang Wang,^b Weimin Wang^a

^aDivision of Molecular Biology, Office of Applied Research and Safety Assessment, Center for Food Safety and Applied Nutrition, U.S. Food and Drug Administration, Laurel, Maryland, USA ^bTranslational Science, MedImmune, Gaithersburg, Maryland, USA

ABSTRACT We report here the complete genome sequence of a GII.6 norovirus strain detected in a clinical fecal specimen from the United States. The virus genome has a length of 7,547 bp and a GC content of 50.1%. Complete norovirus genotyping of the full-genome sequence identified the virus genotype as GII.P6_GII.6.

uman norovirus is the leading causative agent of outbreaks and sporadic cases of acute nonbacterial gastroenteritis across all age groups around the globe, with an estimated prevalence of 20% in cases of acute gastroenteritis in industrialized countries (1). A highly infectious viral pathogen, norovirus can be transmitted through direct contact with infected individuals, aerosol contamination of projectile vomit, and contaminated environmental surfaces or through the consumption of contaminated food and water. Norovirus has been recognized worldwide as the main cause of associated foodborne diseases (2). Noroviruses are a group of genetically diverse viruses that belong to the family Caliciviridae, having a single-stranded positive-sense ~7.6-kb RNA genome in 3 open reading frames (ORFs). ORF1 codes for nonstructural proteins, including RNA-dependent RNA polymerase (RdRp), ORF2 codes for a viral major capsid protein, and ORF3 codes for a minor structural protein. The viruses can be classified into at least 6 genogroups (GI to GVI) based on sequence differences in the major capsid protein (3), with viruses from GI, GII, and GIV known to infect humans. The genogroups are further subdivided into more than 30 genotypes (genetic clusters) (4), of which Gll.6, Gll.3, Gll.12, Gl.3, Gl.6, and Gl.7 have been most often associated with foodborne outbreaks in the United States (5). We previously reported DNA microarray-based detection of a GII.6 norovirus from a clinical fecal sample in the United States (6, 7). To better understand the genomic information and phylogeny of the viral pathogen, we performed whole-genome sequencing of the virus on an Illumina MiSeq platform.

Viral RNA was extracted from clinical fecal materials using the QIAamp viral RNA minikit (Qiagen, USA), as previously described (6). The resulting RNA was reverse transcribed to cDNA and amplified with the Ovation RNA sequencing (RNA-Seq) system version 2 kit (NuGen, USA), according to the manufacturer's protocols. Sequencing libraries were prepared with 1 ng of the amplified products using the Nextera XT library prep kit (Illumina, USA) and sequenced using the Illumina version 2 reagent kit (Illumina). The paired-end sequencing reads were trimmed in CLC Genomics Workbench 9.0.1 (Qiagen), and *de novo* assembly was performed using SPAdes 3.8.1 (8). The *de novo* assembled contigs bigger than 2 kb were searched against the NCBI nonredundant database using BLASTN (Bethesda, MD) to identify norovirus assemblies. The candidate contigs with highest sequence similarities to norovirus sequences were selected for further mapping assembly using Burrows-Wheeler Aligner (BWA) (9). The fully assembled genome was identified as being 7,547 bp in length, with a GC content of 50.1%. The sequencing read depth was 1,445-fold. The viral genome was annotated using the VIGOR program (10, 11) and was predicted to contain three ORFs, of which,

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This is a work of the U.S. Government and is not subject to copyright protection in the United States. Foreign copyrights may apply. Address correspondence to Haifeng Chen, haifeng.chen@fda.hhs.gov. ORF1 encodes the nonstructural polyprotein (1,679 amino acids [aa]), and ORF2 and ORF3 encode the major (547-aa) and minor (258-aa) capsid proteins, respectively. Phylogenetic genotyping analysis of the full genome identified the virus genotype as GII.P6 (RdRp genotype) and GII.6 (capsid genotype) using Norovirus Typing Tool version 2.0 (https://www.rivm.nl/mpf/typingtool/norovirus/). Compared to the full-genome sequences of the phylogenetically closest isolates from Japan (GenBank accession number AB039778) and China (GenBank accession number JX989075), pairwise nucleotide identities were 97.6% and 96.0%, respectively.

Accession number(s). The genome sequence obtained for the strain in this study (Hu/GII.6/186/2005/USA) has been deposited in NCBI GenBank under the accession number HQ169542.

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