

GENOME ANNOUNCEMENT

Genome Sequence of Aggregatibacter actinomycetemcomitans RHAA1, Isolated from a Rhesus Macaque, an Old World Primate

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Aggregatibacter actinomycetemcomitans is implicated in localized aggressive periodontitis. We report the first genome sequence of an A. actinomycetemcomitans strain isolated from an Old World primate.

ggregatibacter actinomycetemcomitans is a Gram-negative oral Abacterium implicated in localized aggressive periodontitis (LAP) (1, 10). Animal models have been used to study LAP (3), and generally, nonhuman primates are recognized as the most appropriate models. Previous studies in our lab have shown that human A. actinomycetemcomitans specifically binds to buccal epithelial cells from humans and Old World but not New World primates (9). A. actinomycetemcomitans strain RHAA1, a serotype b strain, was isolated from a 13-year-old male rhesus macaque (5, 8, 11). The whole genome sequence of A. actinomycetemcomitans RHAA1 was resolved using 454 pyrosequencing technology at a coverage of $75 \times (7)$ (SeqWright Inc., Houston, TX). Assembly was performed using the Newbler assembler, which generated 81 contigs, with most of the bases having a quality score of 64 and above. The contigs were aligned with the genome of reference A. actinomycetemcomitans strain HK1651 (4) (http://www.genome .ou.edu/act.html) using Newbler. Of 34 contig gaps, 29 were closed by PCR and Sanger sequencing. Our efforts to close the remaining 5 gaps were unsuccessful, since mixed chromatograms were obtained when PCR amplicons for these gaps were sequenced, which indicated that these gaps encompassed repetitive regions that are difficult to amplify and sequence. The genome was annotated using the Prokaryotic Genomes Automatic Annotation Pipeline (PGAAP) from NCBI and manually curated. The open reading frames (ORF) were also identified by GLIMMER v 3.02 (2).

The A. actinomycetemcomitans RHAA1 genome has a length of 2,233,070 nucleotides, a GC content of 44.67%, and 2,150 predicted coding sequences. RHAA1 has 48 tRNA genes and 1 rRNA gene as determined by the tool tRNAScan-SE (6). All five genomic islands present in the HK1651 genome, including the cytolethal distending toxin (*cdt*) gene cluster, the tight adherence gene cluster, the O-antigen biosynthesis and transport gene cluster, the leukotoxin gene cluster, and the lipooligosaccharide biosynthesis gene cluster, are present in *A. actinomycetemcomitans* RHAA1. Furthermore, genes coding for major virulence factors of *A. actinomycetemcomitans*, leukotoxin, CDT, fimbriae, poly-*N*-acetylglucosamine (PGA), and PilABCD are present and showed at least 95% identity with the respective sequences from *A. actinomycetemcomitans* HK1651. The leukotoxin operon consists of a non-JP2 type promoter without any deletion.

The genes coding for outer membrane proteins Omp34, Omp64, ApiA, Aae, and EmaA are also found in RHAA1 and were 98% identical to those in *A. actinomycetemcomitans* HK1651. Genes involved in fatty acid and phospholipid metabolism *fadD*,

msbB, *glsX*, and *dgkA* are present. RHAA1 also possesses *tyrR*, *trpB*, and other genes for amino acid biosynthesis. Genes required for cellular processes such as catabolic repression, cell division (i.e., *minC* and *zipA*), and the TCA cycle (i.e., *mdh* and *fumC*) are also present. Genes for iron uptake and utilization, i.e., *tbpA*, *hbpA1*, *hbpA2*, *fecB*, *fecE*, and *fecD*, are present and exhibited 92 to 96% identity to these genes from *A. actinomycetemcomitans* HK1651.

When *A. actinomycetemcomitans* RHAA1 and *Haemophilus influenzae* Rd KW20 were compared, 284 coding sequences were unique to RHAA1 while 179 were unique to KW20. When the genome comparison was made between the *A. actinomycetemcomitans* HK1651 genome and the RHAA1 genome, RHAA1 had 31 unique coding sequences whereas HK1651 had 42.

Nucleotide sequence accession numbers. This whole genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession number AHGR00000000. The version described in this paper is the first version, AHGR01000000.

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