

GENOME ANNOUNCEMENTS

Genome Sequence of Naturally Competent *Aggregatibacter actinomycetemcomitans* Serotype a Strain D7S-1[∇]

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The major clonal lineages of the Gram-negative periodontal pathogen *Aggregatibacter actinomycetemcomitans* include serotype a, b, and c strains. Here, we report the draft genome sequence of a naturally competent serotype a strain, D7S-1, isolated from a patient with aggressive periodontitis.

Gram-negative facultative *Aggregatibacter actinomycetemcomitans* is recognized as a major etiologic agent of aggressive periodontitis (6). Six serotypes have been identified among *A. actinomycetemcomitans* isolates (2, 3); each serotype represents a distinct clonal lineage. The serotype a strain D7S-1 was cultured from a subgingival plaque of an African American female patient diagnosed with generalized aggressive periodontitis. This strain is naturally competent for DNA uptake (7). The complete genome sequencing of the strain was determined by 454 pyrosequencing (4), which achieved 30× coverage. Assembly was performed using the Newbler assembler (454 Life Sciences, Roche Diagnostics Corporation, Branford, CT), which generated 106 large contigs, with 99.95% of the bases having quality scores of 40 and above. The contigs were initially aligned to the genome of the sequenced serotype b strain HK1651 (<http://www.genome.ou.edu/act.html>) and serotype c strain D11S-1 (1) using software written in-house. The selected contig gaps were then closed by primer walking and sequencing of PCR products over the gaps. Subsequently, paired-end sequencing was performed for strain D7S-1, which resulted in 79 large contigs and 32 scaffolds. Inverted PCR was employed from the contig ends to identify potential neighbor contigs. Optical mapping (5) was also used, as an aid for contig alignment. The final gap closure was achieved by primer walking and PCR/sequencing. One gap that we were unable to close was probably due to the presence of a repeat element within this gap. The final sequences of the contig gaps were verified and revised, with all contigs generated by 454 sequencing. The final map was then confirmed with an optical map of an NcoI-restricted D7S-1 genome ([http://expression.washington](http://expression.washington.edu/bumgarnerlab/publications.php)

[.edu/bumgarnerlab/publications.php](http://expression.washington.edu/bumgarnerlab/publications.php)). Additional evidence supporting the contig alignment came from pulsed-field gel electrophoresis analysis of genome fragments of strain D7S-1 generated with I-CeuI. The automated annotation was done using a protocol similar to the annotation engine service at The Institute for Genomic Research/J. Craig Venter Institute, with some local modifications, as described previously (1).

The strain D7S-1 genome contains 2,308,328 nucleotides, a GC content of 44.3%, 2,432 predicted coding sequences, and 53 tRNA and 19 rRNA genes (<http://expression.washington.edu/bumgarnerlab/publications.php>). The distributions of predicted genes based on functional categories were similar among the D7S-1, serotype b HK1651, and serotype c D11S-1 strains (<http://expression.washington.edu/bumgarnerlab/publications.php>). Genomic islands (data not shown) were identified based on annotations for strain HK1651 and manual inspection of contiguous D7S-1-specific DNA regions with G+C bias. Among 9 identified genomic islands, 5 islands (islands D, E, G, H, and I) correspond to the genomic islands in HK1651 (islands 5, 8, 3, 7, and 2, respectively) that carry the leukotoxin gene cluster, the lipooligosaccharide (LOS) biosynthesis enzyme gene cluster, the tight adherence gene cluster, a uncharacterized genomic island, and the cytolethal distending toxin gene cluster, respectively (<http://www.oralgen.lanl.gov/>). The other four genomic islands (islands A, B, C, and F) were unique to strain D7S-1.

Nucleotide sequence accession number. The draft genome sequence of *A. actinomycetemcomitans* strain D7S-1 has been assigned GenBank accession number ADCF00000000. The supporting information can be accessed at <http://expression.washington.edu/bumgarnerlab/publications.php>.

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