Complete Genome Sequence of Acidovorax sp. Strain KKS102, a Polychlorinated-Biphenyl Degrader

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We report the complete genome sequence of Acidovorax sp. strain KKS102, a polychlorinated-biphenyl-degrading strain isolated from a soil sample in Tokyo. The genome contains a single circular 5,196,935-bp chromosome and no plasmids.

KKS102, a betaproteobacterial Acidovorax sp. strain, was isolated from a mixed culture that degrades biphenyl/polychlorinated biphenyls (biphenyl/PCB). KKS102 was not able to grow in minimal medium supplemented with biphenyl/PCB as the sole sources of carbon and energy, but growth was supported by cocultivation with non-biphenyl-degrading Pseudomonas fluorescens KKL101, which was also isolated from the mixed culture (6).

KKS102 has been extensively studied with respect to (i) its metabolic pathway and bph genes, encoding degradation enzymes (2, 3, 5), (ii) its putative symbiosis with KKL101 (4, 6), (iii) the crystal structure of degradation enzymes (13–16), (iv) the forced overexpression of the bph genes through the insertion of a constitutive promoter to create strains with superior degradation activities (12), (v) the alleviation of the toxic effects of biphenyl by degradation (1), (vi) transcriptional regulation involving induction by a pathway intermediate (7, 11) and catabolite repression by organic acids (8), and (vii) the conjugative transfer of a 62-kb integrative and conjugative element carrying all of the bph genes (10).

The KKS102 genome was sequenced by 454 GS-FLX Titanium (Roche) and GAIIx systems (Illumina). For 454 GS-FLX a fragment library was constructed, and 645,167 reads and a total of 191 Mb of data were obtained. For Illumina GAIIx, 101-bp paired-end sequencing was conducted, and 556,324 reads and a total of 162 Mb of data were used after the trimming of reads by over- or underrepresented 17-mers using our original ShortReadManager software. The reads obtained by both systems were assembled using Newbler (Roche) to produce 143 contigs. The finishing was greatly facilitated by our two other original computer programs, GenoFinisher and AceFileViewer. There were 19 types of repeats in the genome; to solve repeat-induced ambiguities, for each type of repeat, a subcontig graph was semiautomatically drawn by GenoFinisher and PCR experiments were designed and performed to reveal pairs of contigs flanking a repeat contig. To close six gaps, we first conducted genome-template sequencing (10) and found three pairs of contigs, each flanking a gap. For the other three gaps, combinatorial PCRs were conducted. AceFileViewer was then employed for (i) the identification of variation bases potentially present in repeat contigs and their subsequent fixation and (ii) the inspection of less-reliable bases and their subsequent manual correction. These three computer programs we developed for finishing the KKS102 genome are available at http://www.ige.tohoku.ac.jp/yohto/af_e/.

The completed sequence was consistent with a macroscale restriction map using I-CeuI, Pmel, and SvaI (data not shown). Sequence annotation was performed using the NCBI Prokaryotic Genomes Automatic Annotation Pipeline (http://www.ncbi.nlm.nih.gov/genomes/static/Pipeline.html), and the resulting annotation was manually inspected with respect to the start codon positions using the Microbial Genome Annotation Pipeline (http://www.migap.org/) as well as another annotation support tool of GenomeMatcher (9).

The complete sequence of the KKS102 genome consisted of a 5,196,935-bp circular chromosome with an average G+C content of 64.0%. A total of 4,744 open reading frames (ORFs), 50 tRNA genes, and 3 rRNA genes were identified. The genome lacks an lysC gene, accounting for its failure to grow in minimal medium.

The complete sequence of KKS102 will aid further studies focusing especially on (i) the elucidation of mechanisms for the catabolite control system and (ii) comparative genomics to gain insight into the evolution of KKS102.

KKS102 is available from the Japan Collection of Microorganisms (KKS102 is deposited under its synonymous name, KKS102S).

Nucleotide sequence accession number. The genome sequence of Acidovorax sp. strain KKS102 has been deposited in NCBI GenBank under accession number CP003872.

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