Genome Sequences of *Alicyclopilus denitrificans* Strains BC and K601TV

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Received 16 March 2011/Accepted 10 June 2011

*Alicyclopilus denitrificans* strain BC and *A. denitrificans* strain K601T degrade cyclic hydrocarbons. These strains have been isolated from a mixture of wastewater treatment plant material and benzene-polluted soil and from a wastewater treatment plant, respectively, suggesting their role in bioremediation of soil and water. Although the strains are phylogenetically closely related, there are some clear physiological differences. The hydrocarbon cyclohexanol, for example, can be degraded by strain K601T but not by strain BC. Furthermore, both strains can use nitrate and oxygen as an electron acceptor, but only strain BC can use chlorate as electron acceptor. To better understand the nitrate and chlorate reduction mechanisms coupled to the oxidation of cyclic compounds, the genomes of *A. denitrificans* strains BC and K601T were sequenced. Here, we report the complete genome sequences of *A. denitrificans* strains BC and K601T.

*Alicyclopilus denitrificans* strain BC and the closely related *A. denitrificans* strain K601T are able to grow with benzene aerobically (6, 8). Furthermore, only strain BC can degrade benzene using chlorate as an electron acceptor (8). During chlorate reduction, molecular oxygen is formed, which might be used by oxygenases to attack benzene (7, 8). Cyclohexanol can be degraded by strain K601T but not by strain BC. Cyclohexanol degradation does not require oxygenases, as was recently reported (4). As comparative genomics of strains BC and K601T may give more insight into anoxic and oxic degradation of cyclic hydrocarbons, the genomes of strains BC and K601T were sequenced.

The genomes of strains BC and K601T were sequenced and assembled at the DOE Joint Genome Institute, and annotation was performed at Oak Ridge National Laboratory (project 4086861). The genomes were based on data from Illumina (2) and 454 (5) technologies, which provided 135× coverage of the genome of strain BC and 416.3× coverage of the genome of strain K601T. Gene annotation was conducted using Prodigal (3).

The total size of the strain BC genome is 4,835,713 bp, slightly smaller than the strain K601T genome (5,070,751 bp). The G+C contents of the genomes are comparable, 67.8% for strain K601T and 67.9% for strain BC. These values are close to those of 66% and 67.7%, respectively, previously determined by conventional techniques (6, 8). Both genomes are comprised of a circular chromosome, which has a size of 4,637,013 bp in strain BC and 4,995,263 bp in strain K601T, and both contain plasmids. Strain BC contains a 78,982-bp plasmid and a 119,718-bp megaplasmid, whereas strain K601T contains one plasmid of 75,488 bp. In total, the genome of strain BC contained 4,545 candidate genes, whereas the genome of strain K601T contained 4,698 gene models.

Remarkably, candidate genes encoding chlorate reductase and chlorite dismutase are localized on the megaplasmid of strain BC, which is absent in strain K601T. This finding explains the predicted horizontal gene transfer of genes involved in chlorate respiration (1). Furthermore, genes coding for a benzene/phenol monooxygenase (BC-BMOa) and a catechol 2,3-dioxygenase (BC-C23O), both postulated to be involved in benzene degradation (8), are localized in one gene cluster in the chromosomes of strain BC and strain K601T. According to our findings, a gene encoding bifunctional hydratase/alcohol dehydrogenase, which is involved in cyclohexanol degradation, was identified in the genome of strain K601T but not in the genome of strain BC.

The genomes of strains BC and K601T contain the blueprints of cyclic hydrocarbon degradation under anoxic and oxic conditions. Interestingly, strain BC can employ aerobic benzene degradation pathways under essentially anaerobic conditions. More detailed genome analysis may lead to the development of methods for application of the strains in *in situ* bioremediation of polluted anaerobic soil sites.

**Nucleotide sequence accession numbers.** The sequences of the chromosomes and plasmids of *A. denitrificans* strains BC and K601T have been deposited in GenBank. For strain BC, the accession...
numbers are CP002449 (chromosome), CP002450 (megaplasmid), and CP002451 (plasmid). For strain K601\textsuperscript{T}, the accession numbers are CP002657 (chromosome) and CP002658 (plasmid).

We gratefully thank the members of the users’ committee of STW project 08053 for discussion and advice.

This research was supported by the Technology Foundation, the Applied Science Division (STW) of the Netherlands Organization for Scientific Research (NWO), project number 08053. H. Junca acknowledges financial support through Colombian Excellence Research Center GeBiX and to contracts 427-2009 and 718-2009 by Colciencias. The work conducted by the U.S. Department of Energy Joint Genome Institute is supported by the Office of Science of the U.S. Department of Energy under contract no. DE-AC02-05CH11231.

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