

# Draft Genome Sequence of *Ralstonia* sp. Strain GA3-3, Isolated from Australian Suburban Soil

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***Ralstonia* sp. strain GA3-3 is a hexachlorocyclohexane (HCH)-degrading bacterial strain isolated from suburban soil in Canberra, Australia. The genome of strain GA3-3 was sequenced to investigate its ability to degrade  $\alpha$ -HCH. Here, we report the annotated genome sequence of this strain.**

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Several bacterial strains were isolated during a hexachlorocyclohexane (HCH) degradation study in Australia. Two strains, including *Ralstonia* sp. strain GA3-3, were chosen to be sequenced based on their HCH degradation phenotype. GA3-3 was isolated from suburban soil with no known history of HCH exposure in Canberra, Australia, after three cycles of enrichment with 50 ppm  $\alpha$ -HCH as the sole carbon source. Further study of GA3-3 revealed that it was able to degrade  $\alpha$ -HCH, albeit incompletely and very slowly compared to other known HCH degraders (1).

The genomic DNA of GA3-3 was prepared using the Qiagen Genomic-tip 20/G kit for bacteria, according to the manufacturer's instructions. Fragments of 500-bp length were then sequenced using Illumina HiSeq 2000 technology at the John Curtin School of Medical Research, Australian National University. The Ray assembler was used to assemble 8,533,778 100-bp paired-end reads using a *k*-mer length of 63 (2). This assembly generated 62 contigs of >500 bp in length, with an  $N_{50}$  value of 193,594 bp. The total size of the assembly was 6.7 Mb, with a G+C content of 66.7%. The paired-end information was able to combine 26 of the contigs into 11 scaffolds, making a total of 47 scaffolds or contigs for the assembly.

Annotation of the genome using the NCBI Prokaryotic Genomes Automatic Annotation Pipeline (PGAAP) predicted 6,174 protein-coding sequences in GA3-3, with 1,439 (23.4%) annotated only as hypothetical proteins. The assembly was also predicted to contain 55 tRNA and 14 rRNA sequences. A BLAST search of the GA3-3 16S rRNA sequence against the NCBI database revealed 99.7% (1,403-nucleotide alignment) identity between GA3-3 and other *Ralstonia* spp.; however, further investigation is needed to determine the true identity of GA3-3.

All previously studied HCH-degrading bacteria share the same pathway for HCH degradation, which requires the *linA-linF* genes (3–6). A BLAST search was performed on

the genome of GA3-3 using the protein sequences encoded by the *linA-linF* genes (accession no. YP\_003545302, BAI96793, YP\_003544005, YP\_003547114, YP\_003547119, and BAI98845, for *linA*, *linB*, *linC*, *linD*, *linE*, and *linF*, respectively) of *Sphingobium japonicum* UT26S from the NCBI database. However, no *linA-linF* genes or their homologues were detected in GA3-3 (1). Another  $\alpha$ -HCH degradation study that was performed using GA3-3 at this stage revealed no degradation activity (S. Pearce, unpublished data). The absence of these genes, coupled with the knowledge that the *lin* genes are normally associated with the transposable insertion element IS6100 (5, 7, 8), may explain the loss of activity since the first degradation experiment.

**Nucleotide sequence accession numbers.** This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession no. [AQPZ00000000](https://www.ncbi.nlm.nih.gov/nuclink/AQPZ00000000). The version described in this paper is version [AQPZ01000000](https://www.ncbi.nlm.nih.gov/nuclink/AQPZ01000000).

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