

Improved Complete Genome Sequence of the Extremely Radioresistant Bacterium *Deinococcus radiodurans* R1 Obtained Using PacBio Single-Molecule Sequencing

Xiaoting Hua,^{a,b} Yuejin Hua^a

Key Laboratory of Chinese Ministry of Agriculture for Nuclear-Agricultural Sciences, Institute of Nuclear-Agricultural Sciences, Zhejiang University, Hangzhou, Zhejiang, People's Republic of China^a; Department of Infectious Diseases, Sir Run Run Shaw Hospital, College of Medicine, Zhejiang University, Hangzhou, Zhejiang, People's Republic of China^b

The genome sequence of *Deinococcus radiodurans* R1 was published in 1999. We resequenced *D. radiodurans* R1 using PacBio and compared the sequence with the published one. Large insertions and single nucleotide polymorphisms (SNPs) were observed among the genome sequences. A more accurate genome sequence will be helpful to studies of *D. radiodurans*.

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Address correspondence to Yuejin Hua, yjhua@zju.edu.cn.

Deinococcus radiodurans R1 is extremely resistant to the lethal effects of ionizing radiation (IR), UV light, oxidation, and desiccation (1). It can survive doses of ionizing radiation of >12,000 Gy, 3,000 times higher than for most vertebrates (2). Different resistance mechanisms have been proposed to explain the extreme radioresistance of the bacterium after its discovery in the 1950s (3). Efficient scavenging of reactive oxygen species and repair of damaged DNA were considered as two of these (4). Over the past decade, many genetic, biochemical, biophysical, and structural studies focused on the DNA repair mechanism of *D. radiodurans* (5–8). These studies were based on the reference sequence of *D. radiodurans*. However, the current genome sequence of *D. radiodurans* R1 was finished in 1999, and contained a number of mistakes in the sequence (9). To fully facilitate studies of *D. radiodurans*, it was necessary to resequence the genome of *D. radiodurans* R1 to provide more accuracy and higher quality.

Here, we present the complete genome sequence of *D. radiodurans* R1, obtained using Pacific BioSciences (PacBio) sequencing technology. Genomic DNA of strain R1 was extracted using a QIAamp DNA minikit (Qiagen, Valencia, CA) following the protocol of the manufacturer. The quality of DNA was determined by gel electrophoresis and a NanoDrop 2000 spectrophotometer (Nano-drop Technologies, Wilmington, DE). After the library construction, the genome was sequenced by the PacBio RS platform. A total of 59,327 polymerase reads with a mean read length of 11,445 bases were generated, which led to a total of 679,027,015 bases with a 176-fold average coverage. *De novo* assembly of the read sequences was performed using continuous long reads following the Hierarchical Genome Assembly Process (HGAP) workflow (PacBio DevNet; Pacific Biosciences) as available in SMRT Analysis v2.3.0. Annotation of *D. radiodurans* R1 was performed using the NCBI PGAAP annotation pipeline and manually checked. The pipeline uses Genemark to predict open reading frames (ORF) and searches against Proteins Clusters. Protein cod-

ing genes were searched against the NCBI RefSeq database using BLASTp.

The genome of *D. radiodurans* R1 is 3,344,765 nucleotides, 66.3% G+C content, and contains two circular chromosomes and two circular plasmids. Among of the 3,212 genes predicted, 3,079 were protein-coding genes. Sixty-two RNAs were also identified. Comparative genome analysis of the genome sequence from this study and the published one was performed with Mauve (10). Large insertions were observed in two chromosomes and two plasmids. Moreover, small insertions and deletions frequently happened in the genome sequence of the bacterium. After mapping the raw sequence data to the previous genome sequence, 92 deletions, 297 insertions, and 188 substitutions were observed. For the DNA repair related gene, frameshifts were detected in the *ssb* gene, which confirmed a previous report. The genome sequence presented here will be helpful for elucidating the radioresistance mechanism in *D. radiodurans*.

Accession number(s). The sequence data for the genome of *D. radiodurans* R1 have been deposited in GenBank under accession numbers [CP015081](https://ncbi.nlm.nih.gov/nucl/CP015081) to [CP015084](https://ncbi.nlm.nih.gov/nucl/CP015084).

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