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Draft Genome Sequence of *Rhodococcus erythropolis* JCM 6824, an Aurachin RE Antibiotic Producer

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***Rhodococcus erythropolis* JCM 6824 is the producer of the quinoline antibiotic aurachin RE. This bacterium also degrades and utilizes some aromatic compounds, such as biphenyl and benzoate. Here, we report the draft genome sequence of this strain.**

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Members of the genus *Rhodococcus* are well known for their prominent ability to degrade or utilize diverse recalcitrant compounds, especially aromatics, such as biphenyls, dioxins, and nitrophenols (1, 2). In terms of this, the number of genes involved in degradation and the whole-genomic DNA sequence of *Rhodococcus* species have been analyzed (3–5). In addition to the favorable characteristics, we have demonstrated that members of the genus *Rhodococcus* are prospective antibiotic producers (6, 7). To date, at least 19 strains of *Rhodococcus* have been shown to have antibiotic-producing properties (6, 8). Of these strains, *Rhodococcus erythropolis* JCM 6824, which was originally isolated as a cholesterol-degrading microorganism (9), produces a quinoline antibiotic, aurachin RE (7). Aurachins are potent antibiotic compounds that exert strong activity against Gram-positive bacteria (7, 10, 11). Although a few antibiotic peptides have been reported so far (12, 13), aurachin RE is the first example of a second metabolism antibiotic isolated from rhodococci. A biosynthesis gene cluster (*rau* genes) of the compound was also identified and investigated in strain JCM 6824 (14). However, none of the genomic DNA sequences of antibiotic-producing rhodococci have been reported to date. To better understand the antibiotic production and other abilities, draft genome sequence analysis of *R. erythropolis* JCM 6824 was performed.

The genome of *R. erythropolis* JCM 6824 was determined using the Illumina GAIIx paired-end technology provided by the Pathogen Genomics Center, National Institute of Infectious Diseases (Tokyo, Japan). This sequencing run yielded 22,830,367 high-quality filtered reads, with 80-bp paired-end sequencing, providing approximately 200× genome coverage. The genome was assembled using the Velvet assembler version 1.1.05 (15). The final assembly consists of 198 scaffolds of 284 contigs containing 7,023,610 bp, with 62.3% G+C content and an N_{50} length of 407,696 bp. The prediction of protein-coding sequences (CDS) and annotation were performed by the Microbial Genome Annotation Pipeline (http://www.migap.org/), which utilizes MetaGeneAnnotator (16), RNAmmer (17), tRNAscan-SE (18), and BLAST (19).

The draft genome sequence of strain JCM 6824 contains 6,718 putative CDSs, 51 tRNAs, and 3 rRNAs. It also contains 18 copies of the putative cytochrome P450 gene, in addition to one gene that was found in the *rau* gene cluster (14, 20). Additionally, it contains putative second metabolism biosynthesis gene clusters, such as nonribosomal peptide synthetase (10 copies), polyketide synthetase (2 copies), and terpene synthetase (2 copies). In addition, it also contains 7 probable aromatic ring-hydroxylating dioxygenase genes and 6 cholesterol oxidase genes, which were estimated to be involved in the initial step of the degradation of these compounds. This genome information may help to understand the function of biosynthesis of second metabolites and also the biodegradation of aromatics and cholesterols in this strain.

Nucleotide sequence accession numbers. The draft genome sequence has been deposited at DDBJ/EMBL/GenBank under accession numbers DF836092 to DF836289. The whole-genome shotgun master numbers are BBLL01000001 to BBLL01000284.

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REFERENCES

1. **Kitagawa W, Miyauchi K, Masai E, Fukuda M.** 2001. Cloning and characterization of benzoate catabolic genes in the Gram-positive polychlorinated biphenyl degrader *Rhodococcus* sp. strain RHA1. J. Bacteriol. **183:**6598–6606. http://dx.doi.org/10.1128/JB.183.22.6598-6606.2001.
2. **Kitagawa W, Kimura N, Kamagata Y.** 2004. A novel *p*-nitrophenol degradation gene cluster from a Gram-positive bacterium, *Rhodococcus opacus* SAO101. J. Bacteriol. **186:**4894–4902. http://dx.doi.org/10.1128/JB.186.15.4894-4902.2004.
3. **McLeod MP, Warren RL, Hsiao WW, Araki N, Myhre M, Fernandes C, Miyazawa D, Wong W, Lillquist AL, Wang D, Dosanjh M, Hara H, Petrescu A, Morin RD, Yang G, Stott JM, Schein JE, Shin H, Smailus D, Siddiqui AS, Marra MA, Jones SJ, Holt R, Brinkman FS, Miyauchi K, Fukuda M, Davies JE, Mohn WW, Eltis LD.** 2006. The complete genome of *Rhodococcus* sp. RHA1 provides insights into a catabolic powerhouse. Proc. Natl. Acad. Sci. U. S. A. **103:**15582–15587. http://dx.doi.org/10.1073/pnas.0607048103.

4. **Kitagawa W, Suzuki A, Hoaki T, Masai E, Fukuda M.** 2001. Multiplicity of aromatic ring hydroxylation dioxygenase genes in a strong PCB degrader, *Rhodococcus* sp. strain RHA1 demonstrated by denaturing gradient gel electrophoresis. Biosci. Biotechnol. Biochem. **65:**1907–1911. http://dx.doi.org/10.1271/bbb.65.1907.
5. **Kulakov LA, Chen S, Allen CC, Larkin MJ.** 2005. Web-type evolution of *Rhodococcus* gene clusters associated with utilization of naphthalene. Appl. Environ. Microbiol. **71:**1754–1764. http://dx.doi.org/10.1128/AEM.71.4.1754-1764.2005.
6. **Kitagawa W, Tamura T.** 2008. Three types of antibiotics produced from *Rhodococcus erythropolis* strains. Microbes Environ. **23:**167–171. http://dx.doi.org/10.1264/jsme2.23.167.
7. **Kitagawa W, Tamura T.** 2008. A quinoline antibiotic from *Rhodococcus erythropolis* JCM 6824. J. Antibiot. **61:**680–682. http://dx.doi.org/10.1038/ja.2008.96.
8. **Nachtigall J, Schneider K, Nicholson G, Goodfellow M, Zinecker H, Imhoff JF, Suessmuth RD, Fiedler H-P.** 2010. Two new aurachins from *Rhodococcus* sp. Acta 2259. J. Antibiot. (Tokyo) **63:**567–569. http://dx.doi.org/10.1038/ja.2010.79.
9. **Watanabe K, Shimizu H, Aihara H, Nakamura R, Suzuki K, Komagata K.** 1986. Isolation and identification of cholesterol-degrading *Rhodococcus* strains from food of animal origin and their cholesterol oxidase activities. J. Gen. Appl. Microbiol. **32:**137–147. http://dx.doi.org/10.2323/jgam.32.137.
10. **Kunze B, Höfle G, Reichenbach H.** 1987. The aurachins, new quinoline antibiotics from myxobacteria: production, physico-chemical and biological properties. J. Antibiot. **40:**258–265. http://dx.doi.org/10.7164/ antibiotics.40.258.
11. **Enomoto M, Kitagawa W, Yasutake Y, Shimizu H.** 2014. Total synthesis of aurachins C, D, and L, and a structurally simplified analog of aurachin C. Biosci. Biotechnol. Biochem. **78:**1324–1327. http://dx.doi.org/10.1080/09168451.2014.918494.
12. **Chiba H, Agematu H, Dobashi K, Yoshioka T.** 1999. Rhodopeptins, novel cyclic tetrapeptides with antifungal activities from *Rhodococcus* sp.II. Structure elucidation. J. Antibiot. **52:**700–709. http://dx.doi.org/10.7164/antibiotics.52.700.
13. **Iwatsuki M, Tomoda H, Uchida R, Gouda H, Hirono S, Ōmura S.** 2006. Lariatins, antimycobacterial peptides produced by *Rhodococcus* sp. K01-B0171, have a lasso structure. J. Am. Chem. Soc. **128:**7486–7491. http://dx.doi.org/10.1021/ja056780z.
14. **Kitagawa W, Ozaki T, Nishioka T, Yasutake Y, Hata M, Nishiyama M, Kuzuyama T, Tamura T.** 2013. Cloning and heterologous expression of the aurachin RE biosynthesis gene cluster afford a new cytochrome P450 for quinoline *N*-hydroxylation. Chembiochem **14:**1085–1093. http://dx.doi.org/10.1002/cbic.201300167.
15. **Zerbino DR, Birney E.** 2008. Velvet: algorithms for *de novo* short read assembly using de Bruijn graphs. Genome Res. **18:**821–829. http://dx.doi.org/10.1101/gr.074492.107.
16. **Noguchi H, Taniguchi T, Itoh T.** 2008. MetaGeneAnnotator: detecting species-specific patterns of ribosomal binding site for precise gene prediction in anonymous prokaryotic and phage genomes. DNA Res. **15:**387–396. http://dx.doi.org/10.1093/dnares/dsn027.
17. **Lagesen K, Hallin P, Rødland EA, Staerfeldt HH, Rognes T, Ussery DW.** 2007. RNAmmer: consistent and rapid annotation of ribosomal RNA genes. Nucleic Acids Res. **35:**3100–3108. http://dx.doi.org/10.1093/nar/gkm160.
18. **Lowe TM, Eddy SR.** 1997. tRNAscan-SE: a program for improved detection of transfer RNA genes in genomic sequence. Nucleic Acids Res. **25:**955–964. http://dx.doi.org/10.1093/nar/25.5.0955.
19. **Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ.** 1990. Basic local alignment search tool. J. Mol. Biol. **215:**403–410. http://dx.doi.org/10.1016/S0022-2836(05)80360-2.
20. **Yasutake Y, Kitagawa W, Hata M, Nishioka T, Ozaki T, Nishiyama M, Kuzuyama T, Tamura T.** 2014. Structure of the quinoline *N*-hydroxylating cytochrome P450 RauA, an essential enzyme that confers antibiotic activity on aurachin alkaloids. FEBS Lett. **588:**105–110. http://dx.doi.org/10.1016/j.febslet.2013.11.016.