Genome sequence of the ethene- and vinyl chloride-oxidizing actinomycete

*Nocardioides* sp. strain JS614

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ABSTRACT

*Nocardioides* sp. strain JS614 grows on ethene and vinyl chloride (VC) as sole carbon and energy sources, and is of interest for bioremediation and biocatalysis. Sequencing the complete genome of JS614 provides insight into the genetic basis of alkene oxidation, supports ongoing research into the physiology and biochemistry of growth on ethene and VC, and provides biomarkers to facilitate detection of VC/ethene-oxidizers in the environment. This is the first genome sequence from the genus *Nocardioides*, and the first genome of a VC/ethene-oxidizing bacterium.

Bacteria in the genus *Nocardioides* (phylum Actinobacteria) are aerobic, non-motile, gram positive rods found in soil. *Nocardioides* strains can metabolise unusual substrates including butane (10), jet fuel (14), phenanthrene (13), p-nitrophenol (31), trinitrophenol (25), atrazine (29), ethene (5) and vinyl chloride (VC) (5).

*Nocardioides* sp., strain JS614 grows on ethene and VC (4)(5), and may be useful for cleaning up VC-contaminated sites (bioremediation) – this is of particular interest due to the carcinogenicity and persistence of VC as a pollutant (15, 26). Strain JS614 is distinguished from other VC degraders (5, 7, 11, 30) by its high activity on VC, and its unusual physiology (20). Strain JS614 can also grow on propene and butene (27), fluoroethene (28) and nicotine (9), and has been proposed as a biocatalyst for production of chiral epoxides (23).

Genome sequencing of JS614 was performed by the U.S. DOE Joint Genome Institute (JGI) and Oak Ridge and Los Alamos National Laboratories (ORNL, LANL) using a Sanger shotgun sequencing approach (8). Libraries were constructed in pUC18c (29,009 reads), pMCL200 (35,995 reads) and pCC1FOS (989 reads), giving genome coverage of 5.2-fold, 6.14-fold and 0.6-fold,
respectively. Reads were compiled using JAZZ (1), and gaps filled by primer walking, PCR, and the
Sequence Finishing Kit (Amersham). Prediction of protein coding sequences was via Glimmer (6)
and Critica (2), with manual confirmation via BLAST against protein databases. Other sequence
features were identified using tRNAscan-RE (18), TMHMM (17), and signalP (3).

The complete JS614 genome (5.3 Mb) consists of a single circular 4,985,871 bp chromosome and one 307,814 bp plasmid (pNOCA01). Although the plasmid was annotated as
circular, pulsed field gel electrophoresis experiments indicated that this element is linear (21). The
average GC content of the chromosome is 71.65%, while that of the plasmid is 68.01%. The
chromosome contains 4645 putative protein-coding genes, 46 tRNAs, and two rRNA operons. The
plasmid contains 256 protein-coding genes. In total, 3457 genes (69%) had function predictions.
The genome sequence confirms that pNOCA01 carries ethene/VC catabolic genes and
coenzyme M biosynthesis genes (16, 19). Like the propene-catabolic genes of Xanthobacter Py2
(NC_009717), the ethene/VC catabolic genes of JS614 have been subject to IS-mediated deletions
and duplications. Plasmid pNOCA01 also carries core metabolic genes (cytochrome C oxidase,
pyruvate dehydrogenase), and a mercury resistance operon. The putative replication gene
(Noca_4704, dnaA homolog) and conjugation coupling gene (Noca_4704, traG homolog) of
pNOCA01 have very high similarity to those of the Arthrobacter plasmid pTC1 (22).

A cluster of 10 putative conjugation genes (Noca_2220-2233) similar to those of linear
plasmids (24) occurs in the chromosome – this could indicate an integrated plasmid or genomic
island. A putative prophage is also present (tape-measure, terminase, and primosome genes;
Noca_550, 555, 570). Other features of interest include a chromate resistance operon (12) and a
respiratory nitrate reductase (Noca_1346-1349) – the latter suggests JS614 could grow anaerobically
by denitrification.
Nucleotide sequence accession numbers. The *Nocardiodes* JS614 sequences have been deposited in GenBank under accession numbers CP000509 and CP000508.

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References


