

Draft Genome Sequence of *Gordonia neofelifaecis* NRRL B-59395, a Cholesterol-Degrading Actinomycete[∇]

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We report a draft sequence of the genome of *Gordonia neofelifaecis* NRRL B-59395, a cholesterol-degrading actinomycete isolated from fresh feces of a clouded leopard (*Neofelis nebulosa*). As predicted, the reported genome contains several gene clusters for cholesterol degradation. This is the second available genome sequence of the family *Gordoniaceae*.

The genus *Gordonia* was originally proposed by Tsukamura (in 1971) and belongs to the mycolic acid group of the *Actinomycetes* (13, 15). In recent years, it has attracted much interest for its ability to degrade xenobiotics, environmental pollutants (1), and recalcitrant natural polymers, such as rubber (9), dibenzothiophene (7), explosive hexogen (14), butyl benzyl phthalates, and cholesterol (4, 10). Quite a number of *Gordonia* species are opportunistic pathogens (6). However, in the genome database, the only genome sequence available at this time is *Gordonia bronchialis* DSM 43247 (5). Recently, our laboratory isolated and characterized *Gordonia neofelifaecis* (NRRL B-59395^T) (11), a novel species of *Gordonia* which shows a high degree of ability to degrade steroidal compounds, and a draft genome sequence of this species is presented here.

The genome sequence was determined using Illumina Genome Analyzer II at the Beijing Genomics Institute (BGI) (Shenzhen, China). Draft assemblies were based on 500-Mb reads. All of the reads provided about 117-fold coverage of the genome. The GAI paired-end reads were assembled into 168 contigs in 47 scaffolds with the SOAPdenovo program. Putative open reading frames encoding more than 30 amino acid residues were predicted using Glimmer 3.0 (3). rRNA genes were identified by RNAmmer (8) and the tRNAscan-SE server (12). The scaffolds were searched against the KEGG and COG (Clusters of Orthologous Groups) databases to annotate the gene descriptions.

The draft genome of *G. neofelifaecis* NRRL B 59395 consists of 4,257,286 bases with a G+C content of 68.62%, and there are 4,032 putative coding sequences with an average length of 951 bp. The coding percentage is 90.3%. There are 46 genes for tRNAs and 5 rRNA loci.

A plethora of genes related to the catabolism of cholesterol were discovered. There are two genes encoding putative 3-ketosteroid- δ -1-dehydrogenase (KSTD), with 60 to 76% identity to orthologs in *R. erythropolis* SQ1; they catalyze the transhydrogenation of 3-keto-4-ene-steroid to 3-keto-1,4-diene-steroid. Three open reading frames encoding 3-ketoster-

oid 9- α -hydroxylase (KshA and KshB) were found. A gene cluster, SCNU_09391 to SCNU_09186, including about 30 genes, was discovered; most of the products of these genes share greatest identity to the ro04482-ro04705 cluster in *Rhodococcus jostii* RHA1 and the rv3492c-rv3574 cluster in *Mycobacterium tuberculosis* H37Rv (16), which encode all of the enzymes necessary to perform one full cycle of β -oxidation and are involved in shortening the cholesterol side chains. Similar to the *igr* locus Rv3545c to Rv3540c in *M. tuberculosis* (2, 16), the second cluster, SCNU_16768 to SCNU_16798, consists of a single operon containing genes for three acyl coenzyme A (acyl-CoA) dehydrogenases, a lipid-transfer protein, and a putative TetR-family transcriptional regulatory protein. A third cluster, SCNU_09181 to SCNU_09146, includes eight genes that are predicted to encode a multicomponent cholesterol uptake system: *mce4ABCDEF* and *supAB* (called the *mce* cluster) (16). The genome sequence of *G. neofelifaecis* and its annotation will provide further insight into the physiology and metabolic potential of *Gordonia*.

Nucleotide sequence accession numbers. This Whole Genome Shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession number AEUD00000000. The version described in this paper is the first version, AEUD01000000.

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REFERENCES

1. Arenskötter, M., D. Broker, and A. Steinbuchel. 2004. Biology of the metabolically diverse genus *Gordonia*. Appl. Environ. Microbiol. **70**:3195–3204.
2. Chang, J. C., et al. 2009. *igr* genes and *Mycobacterium tuberculosis* cholesterol metabolism. J. Bacteriol. **191**:5232–5239.
3. Delcher, A. L., K. A. Bratke, E. C. Powers, and S. L. Salzberg. 2007. Identifying bacterial genes and endosymbiont DNA with Glimmer. Bioinformatics **23**:673–679.
4. Drzyzga, O., L. J. Navarro, L. H. L. Fernández, F. E. García, and J. Perera. 2009. *Gordonia cholesterolivorans* sp. nov., a cholesterol-degrading actinomycete isolated from sewage sludge. Int. J. Syst. Evol. Microbiol. **59**:1011–1015.
5. Ivanova, N., et al. 2010. Complete genome sequence of *Gordonia bronchialis* type strain (3410^T). Stand. Genomic Sci. **2**:19–28.
6. Kageyama, A., et al. 2006. *Gordonia araii* sp. nov. and *Gordonia effusa* sp. nov., isolated from patients in Japan. Int. J. Syst. Evol. Microbiol. **56**:1817–1821.
7. Kim, S. B., et al. 2000. *Gordonia amicalis* sp. nov., a novel dibenzothiophene-desulphurizing actinomycete. Int. J. Syst. Evol. Microbiol. **50**:2031–2036.

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8. Lagesen, K., et al. 2007. RNAmmer: consistent and rapid annotation of rRNA genes. *Nucleic Acids Res.* **35**:3100–3108.
9. Linos, A., et al. 2002. *Gordonia westfalica* sp. nov., a novel rubber-degrading actinomycete. *Int. J. Syst. Evol. Microbiol.* **52**:1133–1139.
10. Liu, Y., et al. 2011. Efficient biotransformation of cholesterol to androsta-1,4-diene-3, 17-dione by a newly isolated actinomycete *Gordonia neofelifaecis*. *World J. Microbiol. Biotechnol.* **27**:759–765.
11. Liu, Y. C., et al. 2011. *Gordonia neofelifaecis* sp. nov., a cholesterol side-chain cleaving actinomycete isolated from the faeces of *Neofelis nebulosa*. *Int. J. Syst. Evol. Microbiol.* **61**:165–169.
12. Lowe, T. M., and S. R. Eddy. 1997. tRNAscan-SE: a program for improved detection of tRNA genes in genomic sequence. *Nucleic Acids Res.* **25**:955–964.
13. Stackebrandt, E., J. Smida, and M. D. Collins. 1988. Evidence of phylogenetic heterogeneity within the genus *Rhodococcus*: revival of the genus *Gordonia* (Tsukamura). *J. Gen. Appl. Microbiol.* **34**:341–348.
14. Thompson, K. T., F. H. Crocker, and H. L. Fredrickson. 2005. Mineralization of the cyclic nitramine explosive hexahydro-1,3,5-trinitro-1,3,5-triazine by *Gordonia* and *Williamsia* spp. *Appl. Environ. Microbiol.* **71**:8265–8272.
15. Tsukamura, M. 1971. Proposal of a new genus, *Gordonia*, for slightly acid-fast organisms occurring in sputa of patients with pulmonary disease and in soil. *J. Gen. Microbiol.* **68**:15–26.
16. Van der Geize, R., et al. 2007. A gene cluster encoding cholesterol catabolism in a soil actinomycete provides insight into *Mycobacterium tuberculosis* survival in macrophages. *Proc. Natl. Acad. Sci. U. S. A.* **104**:1947–1952.