



Complete Genome Sequence of *Rothia mucilaginosa* Strain NUM-Rm6536, Isolated from a Human Oral Cavity

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Here, we present the complete genome sequence of *Rothia mucilaginosa* NUM-Rm6536, a strain isolated from the tongue plaque of a healthy human adult. This strain is amenable to genetic manipulation by transformation and so provides a useful foundation for more detailed investigation of this species.

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othia mucilaginosa, previously known as Stomatococcus muci*laginosus*, is a resident of the human oral cavity and upper respiratory tract (1). Although several studies have suggested that R. mucilaginosa comprises one member of a healthy oral microbiota (2-4), there have been an increasing number of reports on infections caused by this organism, especially among immunocompromised patients (5, 6). More recently, this organism was reported as a species isolated from sputum of cystic fibrosis (CF) patients and defined as a new emerging CF pathogen (7). We previously determined the complete genomic sequence of R. mucilaginosa DY-18, a clinical isolate from a persistent apical periodontitis lesion (DDBJ/EMBL/GenBank accession no. AP011540) (8). The genome of DY-18 encodes two sigma factors, one of which is an extracytoplasmic function (ECF) sigma factor markedly up-regulated under disulfide stress (9), but this strain is not amenable to genetic transformation. Therefore, we isolated a transformable strain of R. mucilaginosa (NUM-Rm6536) from tongue plaque of a healthy human adult (our unpublished data) (10). The aim of the present study is to convey the full genome sequence of NUM-Rm6536.

Total bacterial DNA of NUM-Rm6536 was extracted from an overnight culture using a Nucleo Spin Tissue kit (Macherey-Nagel). A 20-kb SMRTbell library was prepared, and the genome was sequenced using a PacBio RS II system (Pacific Biosciences) on a singlemolecule real-time (SMRT) cell using PacBio P6-C4 chemistry.

De novo assembly of 129,310 reads with a mean length of 6,659 bp using the hierarchical genome assembly process (HGAP) algorithm in SMRT Analysis software version 2.3 (11) revealed a closed circular chromosome 2,292,716-bp in size with average coverage of 222.27×. The genome has a G+C content of 59.56%. The genome was then annotated using RAST (Rapid Annotation using Subsystem Technology) software version 2.0 (12), which successfully identified 1,762 coding sequences, as well as 59 RNA sequences. Of these, 45% of the annotated coding sequences fell within 280 subsystems available in the RAST database. The annotated data set presented here should augment future study of this organism in addition to providing resources for genetic manipulation.

Nucleotide sequence accession number. The genome sequence of *Rothia mucilaginosa* strain NUM-Rm6536 has been deposited in the DDBJ/EMBL/GenBank database under accession number AP014938.

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REFERENCES

- Collins MD, Hutson RA, Båverud V, Falsen E. 2000. Characterization of a *Rothia*-like organism from a mouse: description of *Rothia nasimurium* sp. nov. and reclassification of *Stomatococcus mucilaginosus* as *Rothia mucilaginosa* comb. nov. Int J Syst Evol Microbiol 50:1247–1251. http:// dx.doi.org/10.1099/00207713-50-3-1247.
- Kazor CE, Mitchell PM, Lee AM, Stokes LN, Loesche WJ, Dewhirst FE, Paster BJ. 2003. Diversity of bacterial populations on the tongue dorsa of patients with halitosis and healthy patients. J Clin Microbiol 41:558–563. http://dx.doi.org/10.1128/JCM.41.2.558-563.2003.
- Colombo AP, Boches SK, Cotton SL, Goodson JM, Kent R, Haffajee AD, Socransky SS, Hasturk H, Van Dyke TE, Dewhirst F, Paster BJ. 2009. Comparisons of subgingival microbial profiles of refractory periodontitis, severe periodontitis, and periodontal health using the human oral microbe identification microarray. J Periodontol 80:1421–1432. http://dx.doi.org/10.1902/jop.2009.090185.
- Takeshita T, Suzuki N, Nakano Y, Shimazaki Y, Yoneda M, Hirofuji T, Yamashita Y. 2010. Relationship between oral malodor and the global composition of indigenous bacterial populations in saliva. Appl Environ Microbiol 76:2806–2814. http://dx.doi.org/10.1128/AEM.02304-09.
- Ramanan P, Barreto JN, Osmon DR, Tosh PK. 2014. *Rothia* bacteremia: a 10-year experience at Mayo Clinic, Rochester, Minnesota. J Clin Microbiol 52:3184–3189. http://dx.doi.org/10.1128/JCM.01270-14.
- Maraki S, Papadakis IS. 2015. Rothia mucilaginosa pneumonia: a literature review. Infect Dis (Lond) 47:125–129. http://dx.doi.org/10.3109/ 00365548.2014.980843.
- Lim YW, Schmieder R, Haynes M, Furlan M, Matthews TD, Whiteson K, Poole SJ, Hayes CS, Low DA, Maughan H, Edwards R, Conrad D, Rohwer F. 2013. Mechanistic model of *Rothia mucilaginosa* adaptation toward persistence in the CF lung, based on a genome reconstructed from metagenomic data. PLoS One 8: http://dx.doi.org/10.1371/journal.pone.0064285.
- Yamane K, Nambu T, Yamanaka T, Mashimo C, Sugimori C, Leung K, Fukushima H. 2010. Complete genome sequence of *Rothia mucilaginosa* DY-18: a clinical isolate with dense meshwork-like structures from a per-

sistent apical periodontitis lesion. Sequencing 2010:1-6. http://dx.doi.org/10.1155/2010/457236.

- Nambu T, Yamane K, Yamanaka T, Mashimo C, Maruyama H, Yoshida M, Hayashi H, Leung K-P, Fukushima H. 2013. Identification of disulphide stress-responsive extracytoplasmic function sigma factors in *Rothia mucilaginosa*. Arch Oral Biol 58:681–689. http://dx.doi.org/10.1016/ j.archoralbio.2012.10.017.
- Kobayashi T, Uchibori S, Tsuzukibashi O, Goto H, Aida M. 2012. A selective medium for *Rothia mucilaginosa* and its distribution in oral cavities. J Microbiol Methods 91:364–365. http://dx.doi.org/10.1016/ j.mimet.2012.09.011.
- Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeek RA, McNeil LK, Paarmann D, Paczian T, Parrello B, Pusch GD, Reich C, Stevens R, Vassieva O, Vonstein V, Wilke A, Zagnitko O. 2008. The RAST server: Rapid Annotations using Subsystems Technology. BMC Genomics 9:75. http://dx.doi.org/10.1186/1471-2164-9-75.
 Chin C-S, Alexander DH, Marks P, Klammer AA, Drake J, Heiner C,
- Chin C-S, Alexander DH, Marks P, Klammer AA, Drake J, Heiner C, Clum A, Copeland A, Huddleston J, Eichler EE, Turner SW, Korlach J. 2013. Nonhybrid, finished microbial genome assemblies from long-read SMRT sequencing data. Nat Methods 10:563–569. http://dx.doi.org/ 10.1038/nmeth.2474.