

University of Nebraska - Lincoln

DigitalCommons@University of Nebraska - Lincoln

US Army Research

U.S. Department of Defense

2013

Genome Sequence of *Moraxella macacae* 0408225, a Novel Bacterial Species Isolated from a Cynomolgus Macaque with Epistaxis

Jason T. Ladner

U.S. Army Medical Research Institute of Infectious Diseases

Chris A. Whitehouse

U.S. Army Medical Research Institute of Infectious Diseases

Galina I. Koroleva

U.S. Army Medical Research Institute of Infectious Diseases

Gustavo F. Palacios

U.S. Army Medical Research Institute of Infectious Diseases, gustavo.f.palacios.ctr@us.army.mil

Follow this and additional works at: <https://digitalcommons.unl.edu/usarmyresearch>

Ladner, Jason T.; Whitehouse, Chris A.; Koroleva, Galina I.; and Palacios, Gustavo F., "Genome Sequence of *Moraxella macacae* 0408225, a Novel Bacterial Species Isolated from a Cynomolgus Macaque with Epistaxis" (2013). *US Army Research*. 242.

<https://digitalcommons.unl.edu/usarmyresearch/242>

This Article is brought to you for free and open access by the U.S. Department of Defense at DigitalCommons@University of Nebraska - Lincoln. It has been accepted for inclusion in US Army Research by an authorized administrator of DigitalCommons@University of Nebraska - Lincoln.

Genome Sequence of *Moraxella macacae* 0408225, a Novel Bacterial Species Isolated from a Cynomolgus Macaque with Epistaxis

Jason T. Ladner, Chris A. Whitehouse, Galina I. Koroleva, Gustavo F. Palacios

Center for Genomic Sciences, U.S. Army Medical Research Institute of Infectious Diseases (USAMRIID), Fort Detrick, Maryland, USA

***Moraxella macacae* is a recently described bacterial species that has been associated with at least two outbreaks of epistaxis in macaques. Here we present the first genome sequence of this novel species, isolated from a symptomatic cynomolgus macaque at the U.S. Army Medical Research Institute of Infectious Diseases.**

Received 6 December 2012 Accepted 18 December 2012 Published 14 February 2013

Citation Ladner JT, Whitehouse CA, Koroleva GI, Palacios GF. 2013. Genome sequence of *Moraxella macacae* 0408225, a novel bacterial species isolated from a cynomolgus macaque with epistaxis. *Genome Announc.* 1(1):e00188-12. doi:10.1128/genomeA.00188-12.

Copyright © 2013 Ladner et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 3.0 Unported license](http://creativecommons.org/licenses/by/3.0/).

Address correspondence to Gustavo F. Palacios, gustavo.f.palacios.ctr@us.army.mil.

The genus *Moraxella* within the family *Moraxellaceae* contains at least 14 different species isolated from a variety of mammalian hosts, both terrestrial and aquatic. The most important human-pathogenic species is *M. catarrhalis*, a major cause of upper and lower respiratory tract infections, sinusitis, and childhood otitis media (1). *Moraxella macacae* is a recently described species causing epistaxis, or bloody nose syndrome, in both rhesus macaques from the Tulane National Primate Research Center and cynomolgus macaques from the U.S. Army Medical Research Institute of Infectious Diseases (USAMRIID) (2). Previous outbreaks of epistaxis among cynomolgus macaques have been attributed to *Branhamella (Moraxella) catarrhalis* (3) or *Neisseria (Moraxella) catarrhalis* (4) based on biochemical testing. While the results of phenotypic and biochemical testing of *M. macacae* were consistent with those of *M. catarrhalis*, sequence analysis of the 16S rRNA indicated only 88% identity to *M. catarrhalis* (2). In fact, the closest 16S sequence match was to *M. lincolnii*, a human respiratory tract inhabitant, with 90% nucleotide identity (2).

In the present study, *Moraxella macacae* 0408225, a strain isolated from a cynomolgus macaque (*Macaca fascicularis*) at USAMRIID that demonstrated sneezing with clear to serosanguinous nasal discharge, was chosen for use to sequence and annotate the genome. Genomic DNA was purified using the Qiagen DNeasy blood and tissue kit (Valencia, CA). Whole-genome sequencing was done using a combination of the Illumina GA (paired-end 76-bp reads; ~360× coverage), 454 GSFLX (6-kb mate pairs; ~80× coverage), and Pacific Biosciences (PacBio; 6-kb and 10-kb libraries, ~150× coverage each) platforms. An initial assembly was conducted using Ray (5) with the Illumina and 454 sequences, resulting in five high-quality contigs of ≥500 bp. One contig was found to be an extrachromosomal plasmid, and the entire sequence of this plasmid was verified through Sanger sequencing. The PacBio continuous-long-read [CLR] sequences were then leveraged to join the remaining four contigs using AHA (Pacific Biosciences). Finally, small assembly errors were corrected through an iterative process of mapping the Illumina reads onto the final contigs and then creating a new consensus using Bowtie2, Samtools, and custom scripts (6, 7). The final assembly

consists of two chromosomal contigs (1,213,696 and 865,593 bp) and one plasmid (5,375 bp). The genome is estimated to be ~2.08 Mb, with a G+C content of 39.7%. Automated annotation by NCBI's Prokaryotic Genomes Automatic Annotation Pipeline (PGAAP) revealed 12 rRNA genes, 46 tRNA genes, and 1,828 protein-coding sequences (CDS).

The plasmid encodes a replicase, two mobilization proteins (MobA and MobC), and one hypothetical protein but carries no known virulence factors or antibiotic resistance genes. Results from comparative genomic analysis are consistent with the conclusion that *M. macacae* is a distinct species from *M. catarrhalis*. Reciprocal BLASTp analysis (E value, <1e−10) identified 1,188 (65%) putative orthologs between *M. macacae* and *M. catarrhalis*, with an average of 39.1% amino acid divergence. Additionally, based on 16S rRNA sequences, *M. macacae* falls outside of the clade that contains *M. catarrhalis*; it appears to represent a fifth distinct clade within *Moraxellaceae* (8).

Nucleotide sequence accession numbers. This Whole Genome Shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession number [ANIN000000000](http://www.ncbi.nlm.nih.gov/nuccore/ANIN000000000). The version described in this paper is the first version, [ANIN010000000](http://www.ncbi.nlm.nih.gov/nuccore/ANIN010000000).

ACKNOWLEDGMENTS

We thank O. Elliot for his assistance with the data processing.

This work was funded by the Defense Threat Reduction Agency Project no. 1881290.

Opinions, interpretations, conclusions, and recommendations are those of the authors and are not necessarily endorsed by the U.S. Army.

REFERENCES

- de Vries SPW, van Hijum SAFT, Schueler W, Riesbeck K, Hays JP, Hermans PWM, Bootsma HJ. 2010. Genome analysis of *Moraxella catarrhalis* strain RH4, a human respiratory tract pathogen. *J. Bacteriol.* 192:3574–3583.
- Embers ME, Doyle LA, Whitehouse CA, Selby EB, Chappell M, Philipp MT. 2011. Characterization of a *Moraxella* species that causes epistaxis in macaques. *Vet. Microbiol.* 147:367–375.
- Vandewoude SJ, Luzarraga MB. 1991. The role of *Branhamella-catarrhalis* in the bloody-nose syndrome of Cynomolgus macaques. *Lab. Anim. Sci.* 41:401–406.

4. Olson LC, Palotay JL. 1983. Epistaxis and bullae in cynomolgus macaques (*macaca-fascicularis*). *Lab. Anim. Sci.* 33:377–379.
5. Boisvert S, Laviolette F, Corbeil J. 2010. Ray: simultaneous assembly of reads from a mix of high-throughput sequencing technologies. *J. Comput. Biol.* 17:1519–1533.
6. Langmead B, Salzberg SL. 2012. Fast gapped-read alignment with bowtie 2. *Nat. Methods* 9:357–U354.
7. Li H, Handsaker B, Wysoker A, Fennell T, Ruan J, Homer N, Marth G, Abecasis G, Durbin R, 1000 Genome Project Data Processing Subgroup. 2009. The sequence alignment/map format and SAMtools. *Bioinformatics* 25:2078–2079.
8. Pettersson B, Kodjo A, Ronaghi M, Uhlén M, Tønjum T. 1998. Phylogeny of the family Moraxellaceae by 16S rDNA sequence analysis, with special emphasis on differentiation of *Moraxella* species. *Int. J. Syst. Bacteriol.* 48:75–89.