



Draft Genome Sequences of a Novel Lineage of *Armatimonadetes* Recovered from Japanese Hot Springs

Lewis M. Ward,^a Shawn E. McGlynn,^b Woodward W. Fischer^a

Division of Geological and Planetary Sciences, California Institute of Technology, Pasadena, California, USA^a;
Earth-Life Science Institute, Tokyo Institute of Technology, Meguro-ko, Japan^b

ABSTRACT Here, we report two draft genome sequences from a novel lineage within the *Armatimonadetes* phylum recovered from metagenomes sequenced from Japanese hot spring microbial mats. These organisms are aerobic and represent a new lineage related to the characterized *Chthonomonas* and *Fimbriimonas* groups, and they expand the diversity of this enigmatic phylum.

The *Armatimonadetes* phylum (i.e., OP10) has only four isolates and is primarily known from environmental sequences (1). Here, we describe two closely related draft genome sequences of a novel lineage of *Armatimonadetes* recovered from Japanese hot spring metagenomes.

These genomes were recovered as part of a study of Japanese hot spring microbial mats (2). CP1_70 was recovered from sequencing of a cone-forming microbial mat at Nakabusa Onsen in Nagano Prefecture. This spring is sulfidic and alkaline, and was 48°C with pH 8.3 at the point of sampling. JP3_11 was recovered from a microbial mat sample from an iron-rich intertidal hot spring at Jinata Onsen, Tokyo Prefecture. The spring was 46°C and pH 6.5 at the location of sampling.

Samples (~0.25 cm³) were collected using sterile forceps. DNA was preserved in the field following cell lysis using Zymo TerraLyzer BashingBead matrix and Xpedition lysis buffer. Cells were disrupted by attaching sample tubes to the blade of a cordless reciprocating saw and operating for 1 min. DNA was purified with a Zymo soil/fecal DNA extraction kit (Zymo Research, Irvine, CA) and quantified with a Qubit 3.0 fluorimeter (Life Technologies, Inc., Carlsbad, CA). DNA was submitted to SeqMatic LLC (Fremont, CA) for library preparation and sequencing by Illumina HiSeq. Sequences were assembled with MegaHit version 1.02 (3) and binned using MetaWatt version 3.5.2 (4). Completeness and contamination of bins were estimated using CheckM (5).

The CP1_70 draft genome is 2.45 Mb, has 59.7% GC content, is made up of 470 contigs, and contains 2,176 coding sequences and 47 RNAs. The JP3_11 genome is 2.60 Mb, has 59.4% GC content, is made up of 359 contigs, and contains 2,313 coding sequences and 50 RNAs. Based on an analysis of the single-copy marker genes by CheckM, the CP1_70 draft genome is estimated to be 91.67% complete, and that of JP3_11 is estimated to be 89.35% complete. These genomes contain identical 16S genes; the most similar 16S sequence to those from a cultured organism is only 82% identity to *Acidothermus cellulolyticus*.

Both genomes described here contain genes for aerobic respiration, including alternative complex III (ACIII), an A family heme copper oxidoreductase (HCO), and a B family HCO. The phylogeny of ACIII and B family HCO genes in the *Armatimonadetes* and closely related *Chloroflexi* largely concur with the organismal tree based on conserved ribosomal proteins; consequently, aerobic respiration may represent a vertically inherited ancestral metabolic trait. While members of the *Chloroflexi* lack an outer membrane and associated genes, such as those involved in lipopolysaccharide synthe-

Received 3 July 2017 Accepted 23 August 2017 Published 5 October 2017

Citation Ward LM, McGlynn SE, Fischer WW. 2017. Draft genome sequences of a novel lineage of *Armatimonadetes* recovered from Japanese hot springs. *Genome Announc* 5: e00820-17. <https://doi.org/10.1128/genomeA.00820-17>.

Copyright © 2017 Ward et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Lewis M. Ward, lward@caltech.edu.

sis (e.g., reference 6, and L. M. Ward, J. Hemp, P. M. Shih, S. E. McGlynn, and W. W. Fischer, unpublished data), CP1_70 and JP3_11 contain genes coding for lipopolysaccharide (LPS) synthesis and outer membrane proteins, such as BamA. This suggests that these *Armatimonadetes* are diderm, and that the unusual membrane architecture of the *Chloroflexi* phylum may be a derived trait, a pattern consistent with recent observations from the *Firmicutes* (7).

Accession number(s). This whole-genome shotgun project was deposited in DDBJ/EMBL/GenBank under the accession numbers [NKP000000000](#) (CP1_70) and [NKPV000000000](#) (JP3_11).

ACKNOWLEDGMENTS

L.M.W. acknowledges helpful support from NASA NESSF (grant NNX16AP39H), NSF (grant OISE 1639454), NSF GROW (grant DGE 1144469, the Lewis and Clark Fund for Exploration and Field Research in Astrobiology, and the ELSI Origins Network. S.E.M. acknowledges support from MEXT KAKENHI grant-in-aid for challenging exploratory research (grant award 15K14608). W.W.F. is grateful for the support of NASA Exobiology award NNX16AJ57G, the David and Lucile Packard Foundation, and a Stanford University Blaustein Fellowship.

We thank Katsumi Matsuura and the Environmental Microbiology Laboratory at Tokyo Metropolitan University for laboratory support.

REFERENCES

1. Lee KCY, Dunfield PF, Stott MB. 2014. The phylum *Armatimonadetes*, p 447–458. *In* The prokaryotes. Springer, Berlin, Germany.
2. Ward LM. 2017. Microbial evolution and the rise of oxygen: the roles of contingency and context in shaping the biosphere through time. Ph.D. dissertation. California Institute of Technology, Pasadena, CA. <https://doi.org/10.7907/Z9BZ642S>.
3. Li D, Luo R, Liu CM, Leung CM, Ting HF, Sadakane K, Yamashita H, Lam TW. 2016. MEGAHIT v1.0: a fast and scalable metagenome assembler driven by advanced methodologies and community practices. *Methods* 102: 3–11. <https://doi.org/10.1016/j.ymeth.2016.02.020>.
4. Strous M, Kraft B, Bisdorf R, Tegetmeyer HE. 2012. The binning of metagenomic contigs for microbial physiology of mixed cultures. *Front Microbiol* 3:410. <https://doi.org/10.3389/fmicb.2012.00410>.
5. Parks DH, Imelfort M, Skennerton CT, Hugenholtz P, Tyson GW. 2015. CheckM: assessing the quality of microbial genomes recovered from isolates, single cells, and metagenomes. *Genome Res* 25:1043–1055. <https://doi.org/10.1101/gr.186072.114>.
6. Sutcliffe IC. 2011. Cell envelope architecture in the *Chloroflexi*: a shifting frontline in a phylogenetic turf war. *Environ Microbiol* 13:279–282. <https://doi.org/10.1111/j.1462-2920.2010.02339.x>.
7. Tocheva EI, Ortega DR, Jensen GJ. 2016. Sporulation, bacterial cell envelopes and the origin of life. *Nat Rev Microbiol* 14:535–542. <http://www.nature.com/nrmicro/journal/v14/n8/full/nrmicro.2016.85.html>.