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## Draft Genome Sequence of the Anaerobic Ammonium-Oxidizing Bacterium "*Candidatus* Brocadia sp. 40"

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The anaerobic ammonium-oxidizing (anammox) bacterium "*Candidatus* Brocadia sp. 40" demonstrated the fastest growth rate compared to others in this taxon. Here, we report the 2.93-Mb draft genome sequence of this bacterium, which has 2,565 gene-coding regions, 41 tRNAs, and a single *rrn* operon.

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naerobic ammonium-oxidizing (anammox) bacteria have the ability to convert  $NH_4^+$  to dinitrogen gas using  $NO_2^-$  as an electron acceptor and were first discovered in a bench-scale nitrifying reactor (1). Anammox bacteria were characterized as chemolithoautotrophs and are affiliated with the phylum Planctomycetes (2). Five candidate genera, "Candidatus Kuenenia," "Ca. Brocadia," "Ca. Anammoxoglobus," "Ca. Jettenia," and "Ca. Scalindua," have been proposed in this bacterial lineage to date. Although no pure culture is currently available, several monospecies enrichment cultures have been obtained for anammox bacteria. Anammox-based technologies are widely applied for the treatment of NH<sub>4</sub><sup>+</sup>-rich wastewater (3). However, the application window of this energy- and resource-efficient process is restricted due to the low growth rate of anammox bacteria (4, 5). Recently, "Ca. Brocadia sp. 40" demonstrated a maximum specific growth rate ( $\mu_{\rm max}$ ) of 0.33 days<sup>-1</sup> (6), which is four times higher than the highest value reported so far for anammox bacteria. Here, we describe a high-quality "Ca. Brocadia sp. 40" genome recovered from shotgun metagenomic sequencing of an enrichment culture.

Initially, an anammox bacterium affiliated with "*Ca.* Brocadia sp. 40" was enriched from brewery wastewater sludge in a fixedbed anammox reactor (7). Subsequently, "*Ca.* Brocadia sp. 40" cells were further enriched in a membrane bioreactor. Genomic DNA was extracted using a DNA extraction kit (FastDNA SPIN kit for soil, MP Biomedicals). The library was prepared using a TruSeq Nano DNA library preparation kit (Illumina) and sequenced using a HiSeq 4000 platform (Illumina). Reads were quality-trimmed using Trimmomatic version 0.35 (8) and assembled using SPAdes version 3.9.0 (9). The "*Ca.* Brocadia sp. 40" genome was binned based on coverage and genomic properties using MetaBAT version 0.30.3 (10) as previously described (11, 12).

A 2.93-Mb genome sequence comprising 122 contigs (42.7% GC content) was obtained, and 2,565 gene-coding regions, 41 tRNAs, and a single *rrn* operon were annotated. The genome is 93% complete based on an analysis with CheckM version 1.0.5

(13). The genome size of "*Ca.* Brocadia sp. 40" is smaller and encodes less genes than other anammox species. The nearest evolutionary neighbor, "*Ca.* Brocadia fulgida" (96% 16S rRNA gene sequence similarity), has a genome size of 3.55 Mb (14). This suggests that a smaller genome size is potentially associated with the higher growth of "*Ca.* Brocadia sp. 40" as reported elsewhere (15). The "*Ca.* Brocadia sp. 40" genome encodes gene clusters for core anammox metabolism. However, similar to "*Ca.* Brocadia sinica" (16), conventional nitrite reductase genes (*nirS* and *nirK*) were missing in the "*Ca.* Brocadia sp. 40" genome. It was concluded that anammox bacteria related to the "*Ca.* Brocadia" genus employ an unknown nitrite reductase, which should be further investigated.

Accession number(s). The "*Ca.* Brocadia sp. 40" draft genome was deposited at DDBJ/ENA/GenBank under the accession number MJUW00000000.

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#### REFERENCES

- Mulder A, Graaf AA, Robertson LA, Kuenen JG. 1995. Anaerobic ammonium oxidation discovered in a denitrifying fluidized Bed reactor. FEMS Microbiol Ecol 16:177–183. http://dx.doi.org/10.1111/j.1574 -6941.1995.tb00281.x.
- Strous M, Fuerst JA, Kramer EH, Logemann S, Muyzer G, van de Pas-Schoonen KT, Webb R, Kuenen JG, Jetten MS. 1999. Missing lithotroph identified as new planctomycete. Nature 400:446–449. http:// dx.doi.org/10.1038/22749.
- Ali M, Okabe S. 2015. Anammox-based technologies for nitrogen removal: advances in process start-up and remaining issues. Chemosphere 141: 144–153. http://dx.doi.org/10.1016/j.chemosphere.2015.06.094.
- 4. Strous M, Kuenen JG, Jetten MS. 1999. Key physiology of anaerobic ammonium oxidation. Appl Environ Microbiol 65:3248–3250.
- Ali M, Oshiki M, Awata T, Isobe K, Kimura Z, Yoshikawa H, Hira D, Kindaichi T, Satoh H, Fujii T, Okabe S. 2015. Physiological characterization of anaerobic ammonium oxidizing bacterium "*Candidatus* Jette-

nia caeni. Environ Microbiol 17:2172–2189. http://dx.doi.org/10.1111/ 1462-2920.12674.

- Lotti T, Kleerebezem R, Abelleira-Pereira JM, Abbas B, van Loosdrecht MC. 2015. Faster through training: the anammox case. Water Res 81: 261–268. http://dx.doi.org/10.1016/j.watres.2015.06.001.
- Okamoto H, Kawamura K, Nishiyama T, Fujii T, Furukawa K. 2013. Development of a fixed-bed anammox reactor with high treatment potential. Biodegradation 24:99–110. http://dx.doi.org/10.1007/s10532-012 -9561-x.
- Bolger AM, Lohse M, Usadel B, Planck M, Plant M, Mühlenberg A. 2014. Trimmomatic: a flexible trimmer for Illumina sequence data. Bioinformatics 30:2114–2120. http://dx.doi.org/10.1093/bioinformatics/ btu170.
- Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham S, Prjibelski AD, Pyshkin AV, Sirotkin AV, Vyahhi N, Tesler G, Alekseyev MA, Pevzner PA. 2012. SPAdes: a new genome assembly algorithm and its applications to singlecell sequencing. J Comput Biol 19:455–477. http://dx.doi.org/10.1089/ cmb.2012.0021.
- Kang DD, Froula J, Egan R, Wang Z. 2015. MetaBAT, an efficient tool for accurately reconstructing single genomes from complex microbial communities. PeerJ 3:e1165. http://dx.doi.org/10.7717/peerj.1165.

- Haroon MF, Thompson LR, Parks DH, Hugenholtz P, Stingl U. 2016. A catalogue of 136 microbial draft genomes from Red Sea metagenomes. Sci Data 3:1–6. http://dx.doi.org/10.1038/sdata.2016.50.
- Haroon MF, Thompson LR, Stingl U. 2016. Draft genome sequence of uncultured SAR324 bacterium lautmerah10, binned from a Red Sea metagenome. Genome Announc 4(1):1–2. http://dx.doi.org/10.1128/ genomeA.01711-15.
- 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. http:// dx.doi.org/10.1101/gr.186072.114.
- Gori F, Tringe SG, Kartal B, Machiori E, Jetten MSM, Jetten MS. 2011. The metagenomic basis of anammox metabolism in *Candidatus* 'Brocadia fulgida.' Biochem Soc Trans 39:1799–1804. http://dx.doi.org/10.1042/ BST20110707.
- Kurokawa M, Seno S, Matsuda H, Ying B-W. 2016. Correlation between genome reduction and bacterial growth. DNA Res [Epub ahead of print.] http://dx.doi.org/10.1093/dnares/dsw035.
- Oshiki M, Shinyako-Hata K, Satoh H, Okabe S. 2015. Draft genome sequence of an anaerobic ammonium-oxidizing bacterium, "*Candidatus* Brocadia sinica." Genome Announc 3(2):3–4. http://dx.doi.org/10.1128/ genomeA.00267-15.