





## 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 genecoding regions, 41 tRNAs, and a single *rrn* operon.

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naerobic ammonium-oxidizing (anammox) bacteria have the ability to convert NH<sub>4</sub><sup>+</sup> to dinitrogen gas using NO<sub>2</sub><sup>-</sup> 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 fixed-bed 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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