



# Genome Sequence of the Estuarine *Synechococcus* sp. Strain NB0720\_010

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**ABSTRACT** Marine *Synechococcus* spp. are unicellular cyanobacteria widely distributed in the world's oceans. We report the complete genome sequence of *Synechococcus* sp. strain NB0720\_010, isolated from Narragansett Bay, Rhode Island. NB0720\_10 has several large (>3,000-amino acid) protein-coding genes that may be important in its interactions with other cells, including grazers in estuarine habitats.

Marine *Synechococcus* spp. are photosynthetic bacteria found in the surface waters of nearly every marine habitat on the planet and are estimated to contribute to ~17% of annual net marine primary productivity (1). While there are many genomes available for open-ocean *Synechococcus* spp. (2, 3), there has been limited genome sequencing of coastal and estuarine isolates (4). The genome sequence of *Synechococcus* sp. strain NB0720\_010, isolated from Narragansett Bay and belonging to the marine *Synechococcus* subcluster 5.2 clade CB5, adds to our knowledge of genomic diversity in nearshore and estuarine *Synechococcus* populations.

Strain NB0720\_010 was isolated from water collected on 28 July 2020 at 10:15 a.m. on an incoming tide in Narragansett Bay from a dock at 41°38'59.5812"N, 71°15'24.2038"W. NB0720\_010 was isolated by pour plating using the methods outlined in reference 5 with modifications. Sample seawater was prefiltered through a 1- $\mu$ m filter, mixed with an equal volume of 2 $\times$  PRO99 medium (5) with 0.64% molten (~33°C) low-melting-point agarose, and immediately poured onto a 0.7% agarose PRO99 base layer. The plate was incubated at 22°C with a 14:10-h light/dark illumination cycle at ~20  $\mu$ E m<sup>-2</sup> s<sup>-1</sup> for several weeks until a single colony was picked and transferred to Pro99 liquid medium. After a few transfers to liquid medium, DNA was extracted from a late-exponential-phase culture using a phenol-chloroform extraction protocol (6, 7).

Illumina shotgun sequencing performed by the Microbial Genome Sequencing Center (Pittsburgh, PA; library preparation using the Nextera XT kit following the manufacturer's directions; sequencer, NextSeq 550) yielded 2,649,963 paired-end 151-bp reads. Default parameters were used for all software unless otherwise specified. The reads were trimmed using Trimmomatic v0.38 (8) with the following settings: ILLUMINACLIP:TruSeq3-PE-2.fa:2:30:10, LEADING:10, TRAILING:10 SLIDINGWINDOW:4:15 MINLEN:50. Long-read Nanopore sequencing was performed in-house using DNA from the same culture but extracted from a different inoculum (library preparation using the SQKLSK-110 ligation kit following the manufacturer's directions; Flongle flow cell R9.4.1), and this yielded 874,048 reads (mean length, 255 bp;  $N_{50}$ , 69,887 bp; base calling using Guppy v5.0.11 + 2b6dbff; Oxford Nanopore). Assembly of the Illumina and Nanopore reads using Unicycler v0.4.9b (9) generated 630 contigs ( $N_{50}$ , 8 kb; total length, 18 Mb). Among these was a 2,410,448-bp circular contig with 63% GC content and 97.1% average nucleotide identity (calculated using FastANI [10]) to estuarine *Synechococcus* sp. strain CB0205 (GenBank accession number [GCA\\_000179255.1](https://www.ncbi.nlm.nih.gov/nuccore/GCA_000179255.1)) (11, 12), belonging to subcluster 5.2 clade CB5, and thus, this contig was deemed to be the complete genome of *Synechococcus* sp. NB0720\_010. It was annotated using the NCBI

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**TABLE 1** Characteristics of the four very large (>3,000-aa) proteins in *Synechococcus* sp. strain NB0720\_010

Locus tag	Protein length (aa)	Similarity to other giant proteins in <i>Synechococcus</i> isolate genomes <sup>a</sup>	Domain description
LY254_04120	10,749	30% identity over 44% of the protein to SynWH8101_0818 (4,083 aa; <a href="#">QBE68408.1</a> ) in <i>Synechococcus</i> sp. strain WH 8101 (16)	Several repeated domains (~100 aa long) with similarity to cadherin-like domains (pfam17803, pfam17892), bacterial Ig <sup>b</sup> domains (pfam17963), and repeats (TIGR01965) found frequently in <i>Vibrio</i> , <i>Colwellia</i> , <i>Bradyrhizobium</i> , and <i>Shewanella</i> (VCBS) members
LY254_03050	4,609	30% identity over 82% of the protein to EVJ50_06065 (11,376 aa; <a href="#">QEY31874.1</a> ) in <i>Synechococcus</i> sp. strain RSCCF101 (17) 36% identity over 82% of the protein to <i>Legionella pneumophila</i> toxin protein RtxA ( <a href="#">STX70670.1</a> )	Several repeated domains (DUF5801, pfam19116)
LY254_05360	3,090	No hits to isolate genomes but several to short (<700-aa) proteins from <i>Synechococcaceae</i> freshwater metagenomic assembled genomes (18)	Several repeated cadherin-like domains (pfam17892) and a C-type lectin-like domain (cd03603)
LY254_11890	3,087	96% identity over 100% of the protein to KJJ24_00540 (3,087 aa; <a href="#">QVV67743.1</a> ) in <i>Synechococcus</i> sp. strain LA31 (Narragansett Bay isolate) (4)	RTX toxin domain repeats (NF033203, NF033943) and an RTX toxin-related domain (COG2931)

<sup>a</sup> Top hit(s) from a *Synechococcus* isolate from a blastp search (13) against NCBI's nonredundant (nr) database with an E value of <1e-5. The lengths and GenBank accession numbers of the protein hits are listed in parentheses.

<sup>b</sup> Ig, immunoglobulin.

Prokaryotic Genome Annotation Pipeline v5.3 (13), yielding 2,530 protein-coding genes, 3 complete rRNA operons (5S, 16S, and 23S), and 46 tRNAs.

NB0720\_010 contains several putative type II antitoxin/toxin protein-coding genes. Most of these have homologs (determined using a blastp search [14]; E value < 1e-10) in other estuarine but not open-ocean *Synechococcus* isolate genomes, consistent with previous observations (15). NB0720\_010 also contains four very large (>3,000 amino-acid [aa]), or "giant," protein-coding genes that all have repeated motifs possessing similarity to cadherin-like or RTX toxin domains (Table 1). Such giant proteins often occur in marine *Synechococcus* genomes (19), including SwmB in *Synechococcus* sp. WH8102 (10,791 aa), which is involved in swimming motility and resisting protistan predation (19–21). Similar to SwmB, we suggest that these giant proteins in NB0720\_010 may be important for defense and/or competition, which may be crucial in estuarine waters, which typically have higher cell densities than open-ocean regions.

**Data availability.** *Synechococcus* sp. NB0720\_10 is available from Nathan A. Ahlgren upon request. The sequence data are available at NCBI under BioProject accession number [PRJNA793027](#), including the raw reads (SRA accession numbers [SRR18042579](#) and [SRR18042580](#)) and the assembled genome (GenBank accession number [CP090898](#)).

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