

# Results of 18S sequencing of full-length 18S rDNA for metabarcoding samples collected during R/V Point Sur cruise PS18-09 in the Western Gulf of Mexico in September 2017

**Website:** <https://www.bco-dmo.org/dataset/922101>

**Data Type:** Cruise Results

**Version:** 1

**Version Date:** 2024-03-14

## Project

» [RAPID: Hurricane Impact on Phytoplankton Community Dynamics and Metabolic Response](#) (HRR)

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## Abstract

This dataset includes results of 18S sequencing of full-length 18S rDNA for metabarcoding samples collected from surface depth at stations 06, 11, 16, 21, SS, and GI during R/V Pt. Sur cruise PS18-09 Leg 1 using Nanopore miniON.

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## Coverage

**Location:** Gulf of Mexico, Texas coast

**Spatial Extent:** N:29.0649 E:-94.9 S:27.8358 W:-96.9874

**Temporal Extent:** 2017-09-23

## Methods & Sampling

On September 23, 2017, on HRR cruise Leg 1 (R/V Point Sur PS18-09), samples were collected at 6 stations (S06, S11, S16, S21, SS, and GI) from the surface depth bottle in a CTD rosette. Triplicate 500-1000 milliliter (mL) samples were filtered and immediately fixed in RNALater. Triplicate samples from each station/depth were extracted with AllPrep DNA/RNA MiniKit (Qiagen, USA) following the manufacturer's instructions. DNA concentration and quality were evaluated using a Nanodrop spectrophotometer (Thermo Fisher Scientific Inc, USA). All the samples extracted for DNA were normalized to 5 nanograms per microliter (ng/ $\mu$ L) concentration for the amplicon library construction.

The full-length 18S rDNA region was amplified using 18S primers (Medlin et al. 1988; Edgar & Theriot 2004). Library construction and amplicon sequencing were performed at the Campbell Lab, Texas A&M University using Oxford Nanopore minION. The output sequencing results as fasta files were deposited in NCBI's GenBank under the project number PRJNA592369.

## Data Processing Description

Fastq files for all six station samples were quality checked using FastQC. Resulting sequences were taxonomically annotated using BLAST search against PR2 database v5.0.0. The BLAST analysis used an assignment approach with similarity was  $\geq 90\%$  and query coverage was  $\geq 1000$  bp against the reference sequence. Any sequence that did not compile with this criterion was not used in this study. The following thresholds for identity with BLAST results were used for taxonomic assignment clustering: genus (95%), family (93%), class (92%), and order (90%). Archaeal and Bacterial sequences were removed from these results.

*Additional tools used are:*

Oxford Nanopore Technologies. Guppy protocol. Available at:

[https://community.nanoporetech.com/docs/prepare/library\\_prep\\_protocols/Guppy-protocol/v/gpb\\_2003\\_v1\\_revax\\_14dec2018/guppy-software-overview](https://community.nanoporetech.com/docs/prepare/library_prep_protocols/Guppy-protocol/v/gpb_2003_v1_revax_14dec2018/guppy-software-overview) [Accessed 21 Aug 2023].

## BCO-DMO Processing Description

- Saved the location table (station number, lat, lon, etc.) submitted as part of the dataset metadata in a separate spreadsheet.
- Imported the 6 original CSV files (provided in zip "HRR\_18S\_BLAST\_results.zip") into the BCO-DMO system.
- Concatenated the 6 CSV files into a single data table.
- Joined the columns from the location table to the primary data table by matching on Station number.
- Saved final file as "922101\_v1\_hrr\_18s\_blast\_results.csv".

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## Data Files

File
<b>922101_v1_hrr_18s_blast_results.csv</b> (Comma Separated Values (.csv), 117.16 MB) MD5:6ec461e1f2a8df4ba226050af4974d59
Primary data file for dataset ID 922101, version 1

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## Related Publications

Edgar, S. M., & Theriot, E. C. (2004). Phylogeny of Aulacoseira (Bacillariophyta) based on molecules and morphology 1. *Journal of Phycology*, 40(4), 772–788. Portico. <https://doi.org/10.1111/j.1529-8817.2004.03126.x>

*Methods*

Medlin, L., Elwood, H. J., Stickel, S., & Sogin, M. L. (1988). The characterization of enzymatically amplified eukaryotic 16S-like rRNA-coding regions. *Gene*, 71(2), 491–499. [https://doi.org/10.1016/0378-1119\(88\)90066-2](https://doi.org/10.1016/0378-1119(88)90066-2)

*Methods*

Oxford Nanopore Technologies. Guppy protocol. Nanopore Community. Available from:

[https://community.nanoporetech.com/docs/prepare/library\\_prep\\_protocols/Guppy-protocol/v/gpb\\_2003\\_v1\\_revax\\_14dec2018/guppy-software-overview](https://community.nanoporetech.com/docs/prepare/library_prep_protocols/Guppy-protocol/v/gpb_2003_v1_revax_14dec2018/guppy-software-overview)

*Software*

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## Related Datasets

### IsRelatedTo

Texas A&M University. HRR, RAPID: Hurricane Impact on Phytoplankton Community Dynamics and Metabolic Response. 2019/11. In: BioProject [Internet]. Bethesda, MD: National Library of Medicine (US), National Center for Biotechnology Information; 2011-. Available from: <http://www.ncbi.nlm.nih.gov/bioproject/PRJNA592369>. NCBI:BioProject: PRJNA592369.

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## Parameters

Parameter	Description	Units
Station	sampling station number	unitless
Leg	cruise leg	unitless
Latitude	latitude of sampling station; positive values = North	decimal degrees
Longitude	longitude of sampling station; negative values = West	decimal degrees
Sequence_ID	OTU sequence generated from the HRR data	unitless
Taxonomy	taxonomic annotation of the O.T.U.	unitless
Similarity	percentage of identical matches	percent
length	alignment length (sequence overlap)	base pairs (bp)
mismatches	number of mismatches	base pairs (bp)
gaps	number of gap openings	unitless
Q_start	start of alignment in query	base pairs (bp)
Q_end	end of alignment in query	base pairs (bp)
R_start	start of alignment in reference sequence	base pairs (bp)
R_end	end of alignment in reference sequence	base pairs (bp)
e_value	number of expected hits of similar quality	unitless
score	Bit-score	unitless
file_name_orig	original name of the csv file for this station+leg	unitless

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## Instruments

<b>Dataset-specific Instrument Name</b>	Oxford Nanopore minION Mk1C
<b>Generic Instrument Name</b>	Automated DNA Sequencer
<b>Dataset-specific Description</b>	Library construction and amplicon sequencing was performed at Campbell Lab, Texas A&M University using Oxford Nanopore minION.
<b>Generic Instrument Description</b>	General term for a laboratory instrument used for deciphering the order of bases in a strand of DNA. Sanger sequencers detect fluorescence from different dyes that are used to identify the A, C, G, and T extension reactions. Contemporary or Pyrosequencer methods are based on detecting the activity of DNA polymerase (a DNA synthesizing enzyme) with another chemoluminescent enzyme. Essentially, the method allows sequencing of a single strand of DNA by synthesizing the complementary strand along it, one base pair at a time, and detecting which base was actually added at each step.

<b>Dataset-specific Instrument Name</b>	CTD rosette bottle
<b>Generic Instrument Name</b>	CTD - profiler
<b>Dataset-specific Description</b>	Samples were collected from the surface depth bottle in a CTD rosette.
<b>Generic Instrument Description</b>	The Conductivity, Temperature, Depth (CTD) unit is an integrated instrument package designed to measure the conductivity, temperature, and pressure (depth) of the water column. The instrument is lowered via cable through the water column. It permits scientists to observe the physical properties in real-time via a conducting cable, which is typically connected to a CTD to a deck unit and computer on a ship. The CTD is often configured with additional optional sensors including fluorometers, transmissometers and/or radiometers. It is often combined with a Rosette of water sampling bottles (e.g. Niskin, GO-FLO) for collecting discrete water samples during the cast. This term applies to profiling CTDs. For fixed CTDs, see <a href="https://www.bco-dmo.org/instrument/869934">https://www.bco-dmo.org/instrument/869934</a> .

<b>Dataset-specific Instrument Name</b>	Nanodrop spectrophotometer
<b>Generic Instrument Name</b>	Spectrophotometer
<b>Dataset-specific Description</b>	DNA concentration and quality were evaluated using a Nanodrop spectrophotometer (Thermo Fisher Scientific Inc, USA).
<b>Generic Instrument Description</b>	An instrument used to measure the relative absorption of electromagnetic radiation of different wavelengths in the near infra-red, visible and ultraviolet wavebands by samples.

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## Deployments

PS1809

<b>Website</b>	<a href="https://www.bco-dmo.org/deployment/784313">https://www.bco-dmo.org/deployment/784313</a>
<b>Platform</b>	R/V Point Sur
<b>Start Date</b>	2017-09-22
<b>End Date</b>	2017-10-03
<b>Description</b>	HRR study with three legs. Chief Scientists: Steve DiMarco (Leg 1); Kristen Thyng (Leg 2); Lisa Campbell (Leg 3). R2R Cruise Page: <a href="https://www.rvdata.us/search/cruise/PS1809">https://www.rvdata.us/search/cruise/PS1809</a>

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## Project Information

### **RAPID: Hurricane Impact on Phytoplankton Community Dynamics and Metabolic Response (HRR)**

**Coverage:** Texas coast

#### *NSF Award Abstract:*

Hurricane Harvey is the strongest hurricane to hit the Texas coast in decades and the resulting tidal surges, flooding and terrestrial runoff have had a severe impact on the coastal ocean. The effects on the phytoplankton, the first link in the food chain, may be unprecedented. To determine how the phytoplankton community will respond to such drastic changes in salinity, nutrient inputs, and potential toxins, immediate and continuous sampling is the only way to fully capture the effects and to identify when conditions return to "normal". An automated, continuous phytoplankton imaging instrument that is deployed on the Texas coast records images of the phytoplankton and permits calculation of the abundance of different species. Together with molecular information on the genes that have been "turned on", or expressed, outcomes of this project will help determine the responses of individual types of phytoplankton. Extreme storms are expected to increase in frequency with future climate change, so the responses identified now will be valuable in predicting how such events will affect these primary producers, which in turn support most of the food webs in marine ecosystems, in the future.

High temporal resolution observations from the Imaging FlowCytobot (IFCB) have revealed that hurricanes in the Gulf of Mexico cause drastic changes in the phytoplankton community structure. The objectives of this RAPID project are: 1) to characterize the dynamics of the phytoplankton species in relation to the environmental variables along the Texas coast; 2) to assess the short and long-term changes in the phytoplankton community; and 3) to identify the strategies of the phytoplankton community for resource acquisition. To accomplish these objectives, this project will utilize IFCB time series to follow phytoplankton community structure during the recovery period from Hurricane Harvey. In addition, two RAPID response cruises (in late September and early October) to sample at 5 sites along a transect from Galveston to Port Aransas, TX. At each station, CTD profiles and water samples from surface and the chlorophyll maximum will be collected for nutrients, carbonate chemistry, and RNA sequencing for metatranscriptomic analysis. Metatranscriptomics can provide an indication of the metabolic strategies employed and functional relationships within the plankton community in response to changes in the environment. The advantage of a metatranscriptomic approach is that the entire molecular response to the environment is captured. So, while the response of phytoplankton to increased nutrient inputs from floodwater runoff is targeted, the responses to other environmental stresses (toxics, hypoxia, acidification) are also captured. Analyses of this time series using multivariate statistical techniques, such as principal component analysis (PCA), and network analysis, a powerful technique for identifying potential interactions among taxa, will provide insights on the environmental factors and metabolic responses structuring the community during the aftermath of the hurricane.

**Related data from the The Texas Observatory for Algal Succession Time-Series (TOAST) can be found at the following:** [https://toast.tamu.edu/timeline?dataset=HRR\\_Cruise](https://toast.tamu.edu/timeline?dataset=HRR_Cruise)

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## Funding

<b>Funding Source</b>	<b>Award</b>
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1760620</a>

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