# Microscopy counts (Beggiatoa-like filaments, Cable bacteria), together with in situ temperature and salinity and surface sediment chlorophyll concentrations, of ex situ sediment cores collected in the Chesapeake Bay during 2017-2018

Website: https://www.bco-dmo.org/dataset/847974

Data Type: Other Field Results

Version: 1

Version Date: 2021-04-07

#### **Proiect**

» <u>Collaborative Research: Probing the Metabolic and Electrical Interactions of Cable Bacteria in Anoxic Sediments</u> (Anoxic Sediment Bacteria Interactions)

Contributors	Affiliation	Role
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#### Abstract

This dataset includes microscopy counts (Beggiatoa-like filaments, Cable bacteria), together with in situ temperature and salinity and surface sediment chlorophyll concentrations, of ex situ sediment cores collected in the Chesapeake Bay during 2017-2018.

# **Table of Contents**

- Coverage
- <u>Dataset Description</u>
  - Methods & Sampling
  - Data Processing Description
- Data Files
- Related Publications
- Parameters
- Instruments
- Project Information
- Funding

# Coverage

Spatial Extent: N:38.55728 E:-76.42794 S:38.55505 W:-76.49402

Temporal Extent: 2017-03-27 - 2018-08-24

# Methods & Sampling

#### Methodology:

Immediately after sediment core retrieval, temperature and salinity of the water overlying sediments were measured with hand-held sensors (Orion Star A329 portable multimeter).

In the lab, surficial sediments were sectioned at 0.5 cm depth increments from which aliquots were collected for microscope enumeration of *Beggiatoa*-like filaments and cable bacteria filaments. *Beggiatoa*-like filament enumeration proceeded by methods adapted from Jorgensen et al. 2010 (doi: 10.1111/j.1574-6941.2010.00918), using inverted light microscopy (Zeiss AxioVert A1) with Utermöhl well slides. Filaments were identified as living *Beggiatoa*-like filaments if they were motile, unpigmented, and contained refractive sulfur granules. The length and diameter of each *Beggiatoa*-like filament encountered was recorded. Data are reported as volumetric density of *Beggiatoa*-like filaments (computed assuming a cylindrical shape), integrated

to 5.0 cm depth.

To enumerate cable bacteria, cells were first detached from sediment particles using methods adapted from Kallmeyer et al. 2008, using density centrifugation with Nycodenz (50% wt/vol), and finally captured on filters (0.2 µm cellulose acetate). The identity of cable bacteria was first confirmed using fluorescent *in situ* hybridization (FISH) with the DSB706 oligoprobe (Schauer et al. 2014) on a subset of samples, and then enumerated following staining by Sybr Green I. A minimum of 200 randomly selected fields were viewed at 630X (Zeiss Axio Imager 2). Data are reported as cumulative length of cable bacteria filaments per volume of sediment, and as cumulative volume of cable bacteria computed assuming a cylindrical shape and cell diameter of 1 micron, each integrated to 4.0 cm depth.

For analysis of chlorophyll *a* (Chl*a*) concentration in surface sediments (0-0.5 cm), pigments were extracted into 90% acetone, applying agitation by gentle sonication, and pooling 2-3 successive extractions. Pigment concentration was determined colorimetrically (ThermoScientific Evolution 60S), applying the empirical formula of (Jeffrey et al. 1975).

#### Sampling and Analytical Procedures:

Replicate sediment cores were collected using a gravity corer (Uwitec; clear PVC liners,  $\emptyset = 8.6$  cm). Immediately after sediment core retrieval, temperature and salinity of the water overlying sediments were measured with hand-held sensors (Orion Star A329 portable multimeter). Cores were then capped, kept in the dark at bottom water temperature in a water bath, and transported back to the laboratory, where they were held in a climate-controlled room. Core sectioning was conducted within 1 day of core retrieval.

# **Data Processing Description**

## **BCO-DMO Processing:**

- changed date format to YYYY-MM-DD;
- converted longitude from positive degrees West to negative degrees East.

[ table of contents | back to top ]

# **Data Files**

# File

microscopy\_counts.csv(Comma Separated Values (.csv), 1.40 KB)

MD5:ec56046bf0ecc9f2a85ca6410fb04bea

Primary data file for dataset ID 847974

[ table of contents | back to top ]

#### **Related Publications**

Jeffrey, S. W., & Humphrey, G. F. (1975). New spectrophotometric equations for determining chlorophylls a, b, c1 and c2 in higher plants, algae and natural phytoplankton. Biochemie Und Physiologie Der Pflanzen, 167(2), 191–194. doi:10.1016/s0015-3796(17)30778-3 <a href="https://doi.org/10.1016/S0015-3796(17)30778-3">https://doi.org/10.1016/S0015-3796(17)30778-3</a> <a href="https://doi.org/10.1016/S0015-3796(17)30778-3">Methods</a>

Jørgensen, B. B., Dunker, R., Grünke, S., & Røy, H. (2010). Filamentous sulfur bacteria, Beggiatoa spp., in arctic marine sediments (Svalbard, 79°N). FEMS Microbiology Ecology, no-no. doi:10.1111/j.1574-6941.2010.00918.x

Methods

Kallmeyer, J., Smith, D. C., Spivack, A. J., & D'Hondt, S. (2008). New cell extraction procedure applied to deep subsurface sediments. Limnology and Oceanography: Methods, 6(6), 236–245. doi:10.4319/lom.2008.6.236 Methods

Schauer, R., Risgaard-Petersen, N., Kjeldsen, K. U., Tataru Bjerg, J. J., B Jørgensen, B., Schramm, A., & Nielsen, L. P. (2014). Succession of cable bacteria and electric currents in marine sediment. The ISME Journal, 8(6), 1314–1322. doi:10.1038/ismej.2013.239

# [ table of contents | back to top ]

# **Parameters**

Parameter	Description	Units
date_local	date of field sampling (local time zone; EST); format: YYYY-MM-DD	unitless
Year	year of field sampling	unitless
Month	month of field sampling	unitless
Day	day of month of field sampling	unitless
site	site of sediment collection	site
lat	latitude	degrees North
lon	longitude	degrees East
site_depth	depth of sampling site	meters (m)
T_degC_BW	Temperature, in situ, of bottom water	degrees Celsius
Salinity_BW	Salinity, in situ, of bottom water	unitless
Begg_Density	Density of Beggiatoa-like filaments, integrated to 5 cm depth	cubic millimeters per square centimeter (mm^3 cm^-2)
DSB_Density_len	Density of cable bacteria filaments, integrated to 4 cm depth, expressed as sum length of filaments	meters per square centimeter (m cm-2)
DSB_Density_vol	Density of cable bacteria filaments, integrated to 4 cm depth, expressed as sum volume of filaments	cubic millimeters per square centimeter (mm^3 cm^-2)
SurfaceChla	Extracted Chla concentration in surface (0-0.5cm depth) sediment	micrograms Chla per gram DW sediment

# [ table of contents | back to top ]

# Instruments

Dataset- specific Instrument Name	Zeiss Axio Imager 2
Generic Instrument Name	Fluorescence Microscope
Generic Instrument Description	Instruments that generate enlarged images of samples using the phenomena of fluorescence and phosphorescence instead of, or in addition to, reflection and absorption of visible light. Includes conventional and inverted instruments.

Dataset- specific Instrument Name	gravity corer (Uwitec)
Generic Instrument Name	Gravity Corer
Generic Instrument Description	The gravity corer allows researchers to sample sediment layers at the bottom of lakes or oceans. The coring device is deployed from the ship and gravity carries it to the seafloor. (http://www.whoi.edu/instruments/viewInstrument.do?id=1079).

Dataset- specific Instrument Name	Zeiss AxioVert A1
Generic Instrument Name	Inverted Microscope
Generic Instrument Description	An inverted microscope is a microscope with its light source and condenser on the top, above the stage pointing down, while the objectives and turret are below the stage pointing up. It was invented in 1850 by J. Lawrence Smith, a faculty member of Tulane University (then named the Medical College of Louisiana). Inverted microscopes are useful for observing living cells or organisms at the bottom of a large container (e.g. a tissue culture flask) under more natural conditions than on a glass slide, as is the case with a conventional microscope. Inverted microscopes are also used in micromanipulation applications where space above the specimen is required for manipulator mechanisms and the microtools they hold, and in metallurgical applications where polished samples can be placed on top of the stage and viewed from underneath using reflecting objectives. The stage on an inverted microscope is usually fixed, and focus is adjusted by moving the objective lens along a vertical axis to bring it closer to or further from the specimen. The focus mechanism typically has a dual concentric knob for coarse and fine adjustment. Depending on the size of the microscope, four to six objective lenses of different magnifications may be fitted to a rotating turret known as a nosepiece. These microscopes may also be fitted with accessories for fitting still and video cameras, fluorescence illumination, confocal scanning and many other applications.

Dataset-specific Instrument Name	Orion Star A329 portable multimeter
Generic Instrument Name	Multi Parameter Portable Meter
	An analytical instrument that can measure multiple parameters, such as pH, EC, TDS, DO and temperature with one device and is portable or hand-held.

Dataset-specific Instrument Name	ThermoScientific Evolution 60S
Generic Instrument Name	Spectrophotometer
Generic Instrument Description	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.

[ table of contents | back to top ]

# Collaborative Research: Probing the Metabolic and Electrical Interactions of Cable Bacteria in Anoxic Sediments (Anoxic Sediment Bacteria Interactions)

**Coverage**: Chesapeake Bay sediments; Mid-Atlantic Coastal sediments

#### NSF Award Abstract:

Marine sediments represent the world's largest repository of stored organic carbon, and understanding how microorganisms break down this carbon is an imperative for understanding global carbon cycling. Yet longstanding questions remain regarding how networks of microorganisms work together to accomplish the complete breakdown of organic carbon in marine sediments. Sediment microbes interact in a myriad of ways that couple their metabolism to the break down of organic carbon, including by sharing products of metabolism. Accumulating evidence further suggests that some microorganisms can interact by transferring electrons directly to other unrelated microorganisms. This ability occurs across diverse microorganisms and appears to be widespread in the biosphere, particularly in anaerobic environments such as marine sediments. This project addresses emerging questions about the identity and metabolic linkages between microorganisms that work together in natural anaerobic marine and estuarine sediments to break down organic carbon. The investigators approach these questions by focusing on the influence of a keystone bacterium on its surrounding microbial community. "Cable bacteria" are a recently discovered group of long filamentous bacteria that act as electrical conductors in aquatic sediments providing a conduit for electrons to commute from deeper sulfidic sediments up to the surface oxygen layer by the process of centimeter-scale electron transport. Since their discovery about 6 years ago, these bacteria have been observed in a wide range of depositional sedimentary environments, often at extremely high cell densities. Where these bacteria are abundant, such as in coastal marine muds, they drive intense localized changes in pH and strongly influence the mineral cycling. This research explores the direct and indirect influence of cable bacteria on the metabolic activity of associated microorganisms. This project also advance the education and training of two early-career investigators, two PhD students, and undergraduate students. The skills and expertise gained from these PhD research projects will enable the students to be competitive in academic pursuits and in bioinformatics and technology applications relevant to private industry. The scientific discoveries emerging from this work is being incorporated into undergraduate and graduate level courses in marine microbial ecology. The research team will reach out to the broader community by hosting public lectures promoting a better understanding of environmental microbial ecology.

The proposed work is to investigate the role of cable bacteria in structuring sediment microbial communities. Due to their growth strategy and morphology, cable bacteria are particularly amenable to experimental manipulation, providing an outstanding opportunity to better understand community interactions among microorganisms in a natural and complex anaerobic environment. The investigators will explore the interactions and relationships between cable bacteria and their associated microbial community by manipulating the growth and activity of cable bacteria and quantifying the resultant microbial community response. Specifically, this project aims to (1) identify microorganisms whose growth is enhanced by cable bacteria, (2) identify metabolic processes linked with cable bacteria activity using metatranscriptomics, (3) test specific metabolic links between sediment microorganisms and cable bacteria activity using a DNA-stable isotope probing (SIP) approach, and (4) visually confirm the identity and quantify key microorganisms associated with cable bacteria using microscopy. As more is learned about the identity and the mechanisms by which microorganisms are metabolically linked in anoxic sediments, we will be better able to understand and make predictions about how microorganisms function in their environment and how they can be utilized in bioengineered systems.

This award reflects NSF's statutory mission and has been deemed worthy of support through evaluation using the Foundation's intellectual merit and broader impacts review criteria.

### [ table of contents | back to top ]

# **Funding**

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1756877
NSF Division of Ocean Sciences (NSF OCE)	OCE-1756851

[ table of contents | back to top ]