

# Genetic accession numbers for viral and bacterial metagenomes from Asteroid sea stars including sea star wasting disease state and collection information from sites worldwide between 2013 and 2016 (Sea Star Microbiology project)

**Website:** <https://www.bco-dmo.org/dataset/728146>

**Data Type:** experimental

**Version:**

**Version Date:** 2018-03-02

## Project

» [Microbial ecology of sea star wasting disease](#) (Sea Star Microbiology)

Contributors	Affiliation	Role
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## Table of Contents

- [Coverage](#)
- [Dataset Description](#)
  - [Methods & Sampling](#)
  - [Data Processing Description](#)
- [Data Files](#)
- [Related Publications](#)
- [Parameters](#)
- [Instruments](#)
- [Deployments](#)
- [Project Information](#)
- [Funding](#)

## Coverage

**Spatial Extent:** N:55.762037 E:151.758016 S:-34.11126 W:-124.713444

**Temporal Extent:** 2013-09-20 - 2016-02-16

## Dataset Description

This dataset contains Asteroid sea star genetic accession numbers at the National Center for Biotechnology Information (NCBI) in addition to information about the sea stars sampled including collection date, location, and notation of sea star wasting disease (SSWD) symptoms. Sea star sampling took place at sites worldwide between 2013 and 2016.

These data were utilized in the following publication (Hewson et al., 2018)

## Methods & Sampling

### For viral metagenomes:

1 g of preserved or frozen tissue was homogenized in 25 mL of 0.02µm filtered PBS in a NutriBullet blender. The homogenate was centrifuged briefly at 3,000 x g to remove large tissue debris and ossicles, and the supernatant filtered through 0.2µm Durapore filters to remove cellular debris. The filtrate was precipitated with PEG-8000 (1 g/ml) overnight. The filtrate was then centrifuged at 22,000 x g for 30 min, the supernatant decanted, and the pellet resuspended in 1 ml 0.02 µm filtered PBS. The resuspension was filtered through a 0.2 µm Acrodisc PES filter. Samples were then treated with DNaseI, RNase One and Benzonase for 3 h.

Nuclease activity was stopped by the addition of x vol EDTA (25 mM). Viral DNA in the purified suspension was extracted using the Zymo Viral DNA kit. The DNA was amplified prior to sequencing using the WGA2 kit V2 (GE Biosciences) before submission to the Cornell Biotechnology Resource Center (BRC) for library preparation and sequencing.

### For bacterial metagenomes:

Samples of asymptomatic asteroid tissues (~ 1cm<sup>2</sup>) were excised from dorsal surfaces and lesioned tissues were excised from symptomatic asteroids, and then homogenized in 10mL of 0.02 µm filtered phosphate buffered saline using the NutriBullet for 1 minute. Homogenates were allowed to settle for 5 min before 1 mL supernatant was removed and placed into sterile microcentrifuge tubes. The homogenate (400 µL) was purified on PERCOLL step gradients (800 µL each step at 70% and 30%) to remove host cells. Gradients were centrifuged at 12,000 x g for 90 min (4°C). The interface between 30% and 70% was determined with reference to a dense *E. coli* culture run on an identical gradient. The interface was removed and DNA extracted using the ZR Bacterial & Fungal DNA kit (Zymo Research) following manufacturer's recommendations. The interface fraction was then targeted for metagenomic library preparation. DNA was extracted from the cell fraction and amplified using the GenomePLEX kit (Sigma Aldrich).

Collection note: The sea star samples were collected by collaborators (samples other than *P. ochraceus* or *P. helianthoides* - these were collected by Ian Hewson) intertidally by hand.

### Data Processing Description

BCO-DMO Data Manager Processing Notes:

- \* added a conventional header with dataset name, PI name, version date
- \* modified parameter names to conform with BCO-DMO naming conventions
- \* converted latitude/longitude in decimal degrees with directional to decimal degrees with South and West as negative values
- \* commas in data changed to semicolons to support csv output\*
- \* added links to NCBI accessions
- \* species name *Parvalusta exigua* changed to *Prvulastra exigua* (accepted name <http://www.marinespecies.org/aphia.php?p=taxdetails&id=459556>) after communication with PI
- \* species name *Mathasterias muizenberg* changed to *Marthasterias glacialis* (accepted name <http://www.marinespecies.org/aphia.php?p=taxdetails&id=123803>) after communication with PI. This may become *Marthasterias glacialis* (subsp. *muizenberg*) in future.

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

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### Data Files

File
<b>SeaStarGenbankAccessions.csv</b> (Comma Separated Values (.csv), 10.64 KB) MD5:5a522caa283d81cd24aff6359dbd2ae7
Primary data file for dataset ID 728146

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

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### Related Publications

Hewson, I., Bistolas, K. S. I., Quijano Cardé, E. M., Button, J. B., Foster, P. J., Flanzenbaum, J. M., ... Lewis, C. K. (2018). Investigating the Complex Association Between Viral Ecology, Environment, and Northeast Pacific Sea Star Wasting. *Frontiers in Marine Science*, 5. doi:[10.3389/fmars.2018.00077](https://doi.org/10.3389/fmars.2018.00077)  
*Results*

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

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## Parameters

Parameter	Description	Units
Accession_number	Link to accession at NCBI	unitless
Library_Name	Name of library	unitless
Library_Description	Description of the library	unitless
Host_Species	Species from which viral/bacterial sequences were obtained	unitless
Disease_State	Sea star wasting disease (SSWD) state (Asymptomatic SSWD-affected)	unitless
Date_of_Collection	Date of host collection in format dd-mmm-yy	unitless
Location_Description	Site location (Country; Site) of host collection	unitless
Latitude	Latitude of host collection	decimal degrees
Longitude	Longitude of host collection	decimal degrees

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

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## Instruments

<b>Dataset-specific Instrument Name</b>	Illumina MiSeq Sequencer
<b>Generic Instrument Name</b>	Automated DNA Sequencer
<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.

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

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## Deployments

### SeaStarMicrobiology\_Hewson

<b>Website</b>	<a href="https://www.bco-dmo.org/deployment/720080">https://www.bco-dmo.org/deployment/720080</a>
<b>Platform</b>	shoreside Alaska

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

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## Project Information

**Microbial ecology of sea star wasting disease (Sea Star Microbiology)**

**Website:** <http://seastarwastingdisease.wordpress.com>

**Coverage:** Salish Sea and Alaskan Waters

Beginning in June 2013 and continuing to present (May 2015), over 20 species of sea stars (Asteroidea, Echinodermata) have been affected by sea star wasting disease (SSWD), affecting populations from central Alaska to Baja California. The disease has led to greatly reduced abundance or disappearance of these keystone predators, which may result in profound alteration to benthic community structure. Recent work has identified the sea star associated densovirus (SSaDV) as the most likely causative agent of the disease. SSaDV is related to densoviruses inhabiting other echinoderms worldwide, and has been present in West Coast asteroid populations for at least 72 years. Hence, there remain significant knowledge gaps in our understanding of how SSaDV actually elicits SSWD symptoms, especially how the echinoderm host, densovirus and microbiome constituents interact. This project will address three major questions: 1) does viral infection change the composition of the sea star microbiome?, 2) what is the variation of viral genomes and their associated virulence?, and 3) does larval dispersal spread the disease between habitats? This project will address these hypotheses through time-course measurements of host, pathogen and associated microorganisms, genome-genome comparisons between historical and contemporary viral strains, and through experiments targeting larvae and juvenile asteroids in aquaria and in nature.

This project will address three fundamental questions relating to Sea Star Wasting Disease (SSWD): 1) How does SSaDV causes SSWD symptoms and how does the disease progress from primary infection through animal mortality; 2) How do current genotypes of SSaDV vary from those present historically, and is virulence related to genome polymorphisms; and 3) Are larvae and juvenile asteroids differentially affected by SSaDV, and are broadcast-spawned bipinnaria a viable mechanism for SSaDV dispersal between distant habitats. The first question will be addressed by experimental inoculation of naïve sea stars with SSaDV, then time-course monitoring of host transcription (i.e. transcriptomics via RNAseq), microbiome composition via 16S rRNA sequencing and quantitative PCR, and viral load and prevalence using quantitative PCR. The second question will be addressed by amplifying the entire genome of SSaDV and related densoviruses, then perform genome-genome comparisons to identify polymorphic DNA in key protein-encoding regions. The third question will be addressed by collecting bipinnaria from plankton at field locations adjacent to spawning asteroid populations, and by performing time-course observations of captive juvenile sea stars and monitoring their bacterial and viral loads using quantitative PCR. This work will be performed primarily in the Salish Sea region, with SSaDV - naïve asteroids collected from Alaskan waters.

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

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## Funding

Funding Source	Award
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1537111</a>

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