## Gene predictions for the complete TetV-1 genome sequence

Website: https://www.bco-dmo.org/dataset/863931 Data Type: Other Field Results Version: 1 Version Date: 2021-10-25

#### Project

» Eating themselves sick? Ecological interactions among a mixotrophic flagellate, its prokaryotic prey, and an ingestible giant virus. (Giant virus ecology)

Contributors	Affiliation	Role
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## Coverage

Spatial Extent: Lat:21.4297 Lon:-157.792 Temporal Extent: 2010-09-02

#### Methods & Sampling

*Note:* The detailed protocols are described in Schvarcz and Steward, 2018.

Shoreline sampling was carried out on September 2nd, 2010 in the coastal surface water of Kāne'ohe Bay, O'ahu, Hawai'i. Seawater (175 L) was filtered (0.8 µm polycarbonate track-etched), and particles in the filtrate concentrated by tangential flow filtration (TFF; 30 kDA molecular weight cut-off, regenerated cellulose). This was then used to challenge a culture of Tetraselmis species isolated previously from the same water. Lysate was serially diluted and used to challenge a new culture for several rounds using a dilution-to-extinction approach to ensure clonality of the virus. A large batch of lysate (40 L of culture) was filtered (0.45 PVDF over a Whatman glass-fiber GF/C). Viruses in the filtrate were concentrated by TFF (30 kDa) to 350 mL volume, further concentrated by polyethylene glycol precipitation, and purified in a CsCl buoyant density gradient. DNA was extracted from the virus peak in the gradient by buffer exchange into TE, treatment with hot SDS and proteinase K, followed by sequential selective precipitation of proteins (salting out), then DNA (alcohol precipitation) using the MasterPure DNA Purification Kit (Epicentre).

DNA was sequenced using PacBio P6-C4 chemistry platform. The complete genome sequence was deposited in GenBank (Clark, 2015) with accession number KY322437

(<u>https://www.ncbi.nlm.nih.gov/nuccore/KY322437</u>). The gene annotations were published as Supplementary Dataset S1 in Schvarcz & Steward, 2018.

#### **Data Processing Description**

Data processing:

Reads were assembled with SPAdes (v3.6.2) which is the St.Petersburg genome assembly algorithm (Bankevich et al., 2012). The assembly was polished using a combination of pbalign v0.2.0.141024 and Quiver v2.0.0, which are part of the PacBio SMRT® Tools (Pacific Biosciences, 2019).

Initial gene prediction was conducted with Prodigal v2.6.3, a gene prediction algorithm (Hyatt et al., 2010), followed by additional steps to identify missed genes. Putative missed genes were identified by querying a larger set of potential open reading frames (ORFs). For example, all potential genes found by Prodigal against the NCBI conserved domain database (CDD). Any hits distinct from Prodigal predictions were further evaluated with searches against:

\* Pfam (<u>http://pfam.xfam.org/;</u> Finn et al., 2016)

\* InterPro (https://www.ebi.ac.uk/interpro/; Jones et al., 2014)

\* NCBI nr databases (blastp, default settings; <u>http://blast.ncbi.nlm.nih.gov/;</u> Camacho et al., 2009).

The annotation of each open reading frame (ORF) was performed manually, after careful evaluation of protein similarity search results from the blastp (nr), CDD, pfam, and InterPro web servers, all using the default settings.

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## **Related Publications**

Bankevich, A., Nurk, S., Antipov, D., Gurevich, A. A., Dvorkin, M., Kulikov, A. S., ... Pevzner, P. A. (2012). SPAdes: A New Genome Assembly Algorithm and Its Applications to Single-Cell Sequencing. Journal of Computational Biology, 19(5), 455–477. doi:<u>10.1089/cmb.2012.0021</u> *Methods* 

Blum, M., Chang, H.-Y., Chuguransky, S., Grego, T., Kandasaamy, S., Mitchell, A., ..., Finn, R. D. (2020). The InterPro protein families and domains database: 20 years on. In Nucleic Acids Research (Vol. 49, Issue D1, pp. D344–D354). Oxford University Press (OUP). https://doi.org/10.1093/nar/gkaa977 https://www.ebi.ac.uk/interpro/ Methods

Camacho, C., Coulouris, G., Avagyan, V., Ma, N., Papadopoulos, J., Bealer, K., & Madden, T. L. (2009). BLAST+: architecture and applications. BMC Bioinformatics, 10(1). doi:<u>10.1186/1471-2105-10-421</u> *Methods* 

Clark, K., Karsch-Mizrachi, I., Lipman, D. J., Ostell, J., & Sayers, E. W. (2015). GenBank. Nucleic Acids Research, 44(D1), D67–D72. doi:<u>10.1093/nar/gkv1276</u> *General* 

Finn, R. D., Coggill, P., Eberhardt, R. Y., Eddy, S. R., Mistry, J., Mitchell, A. L., ... Bateman, A. (2015). The Pfam protein families database: towards a more sustainable future. Nucleic Acids Research, 44(D1), D279–D285. doi:<u>10.1093/nar/gkv1344</u> *Methods* 

Hyatt, D., Chen, G.-L., LoCascio, P. F., Land, M. L., Larimer, F. W., & Hauser, L. J. (2010). Prodigal: prokaryotic gene recognition and translation initiation site identification. BMC Bioinformatics, 11(1). doi:<u>10.1186/1471-2105-11-119</u>

Methods Jones, P., Binns, D., Chang, H.-Y., Fraser, M., Li, W., McAnulla, C., McWilliam, H., Maslen, J., Mitchell, A., Nuka, G., Pesseat, S., Quinn, A.F., Sangrador-Vegas, A., Scheremetijew, M., Yong, S-Y., Lopez, R., and Hunter, S. (2014). InterProScan 5: genome-scale protein function classification. Bioinformatics, 30(9), 1236–1240. doi:10.1093/bioinformatics/btu031 Methods

Nurk, S., Bankevich, A., Antipov, D., Gurevich, A., Korobeynikov, A., Lapidus, A., … Pevzner, P. A. (2013). Assembling Genomes and Mini-metagenomes from Highly Chimeric Reads. Research in Computational Molecular Biology, 158–170. doi:<u>10.1007/978-3-642-37195-0\_13</u> *Methods* 

Pacific Biosciences (2019). SMRT® Tools Reference Guide, P/N 100-939-900 Version 06, (August 2019). Retrieved from <u>https://www.pacb.com/wp-content/uploads/SMRT\_Tools\_Reference\_Guide\_v700.pdf</u>

#### Methods

Schvarcz, C. R., & Steward, G. F. (2018). A giant virus infecting green algae encodes key fermentation genes. Virology, 518, 423–433. doi:<u>10.1016/j.virol.2018.03.010</u> *Methods* 

#### Results

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#### **Related Datasets**

#### Results

Genome [Internet]. Bethesda (MD): National Library of Medicine (US), National Center for Biotechnology Information; [1988] – . Accession No. KY322437.1, Tetraselmis virus 1, complete genome; [cited 2021 Nov 03]. Available from: <u>https://www.ncbi.nlm.nih.gov/nuccore/KY322437.1/</u>

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#### Parameters

Parameters for this dataset have not yet been identified

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#### Instruments

Dataset-specific Instrument Name	PacBio P6-C4 chemistry platform	
Generic Instrument Name	Bioanalyzer	
Dataset-specific Description	DNA was sequenced using PacBio P6-C4 chemistry platform	
Generic Instrument Description	A Bioanalyzer is a laboratory instrument that provides the sizing and quantification of DNA, RNA, and proteins. One example is the Agilent Bioanalyzer 2100.	

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#### **Project Information**

# Eating themselves sick? Ecological interactions among a mixotrophic flagellate, its prokaryotic prey, and an ingestible giant virus. (Giant virus ecology)

**Coverage**: North Pacific Subtropical Gyre - Station ALOHA; and North Pacific tropical embayment, Oahu, HI - Kaneohe Bay

#### NSF Award Abstract:

Phytoplankton support the biological bounty of our seas, so understanding what controls their growth and death is one of the central issues in oceanography. In much of the nutrient-depleted surface waters of the open ocean, the most successful phytoplankton are tiny photosynthetic bacteria known as Prochlorococcus. These bacteria are highly successful competitors for the ocean's limited nutrients and commonly outcompete

larger phytoplankton. Yet, many larger types of phytoplankton persist in the ocean. One reason why this coexistence may occur is that some of the weaker competitors called mixotrophs have evolved a clever alternative strategy best summed up as "If you can't beat them, eat them". In addition to directly competing for nutrients dissolved in the water, these larger phytoplankton can acquire nutrients by consuming and digesting their smaller rivals. The dual ability to photosynthesize and eat competitors has clear advantages, but there can be hidden costs of this intraguild predation strategy. While feeding on Procholorococcus, mixotrophs may also inadvertently ingest giant viruses that are so large they are mistaken for food. Infection is often fatal. Mixotrophy and viral infection are ubiguitous in the ocean; however these processes are often understudied and missing from traditional models of marine food webs that generally consider photosynthesis and predation independently. In this project, the interactions among a common mixotroph (Florenciella), its prey (Prochlorococcus), and a virus that infects the mixotroph (FloV1) will be studied in the lab and field. This research will also help quide the development of a cohesive mixotroph-virus-prey trophic model. Improving these trophic models to account for more complex processes could fundamentally change our understanding of marine trophic dynamics. The project will directly support the training of a post-doc, graduate and undergraduate student in inter-disciplinary science that includes field, lab, and modeling activities. The project will support a major component of the graduate student's dissertation and the progressive training of an undergraduate student, culminating in an independent project. The concepts of mixotrophy and viral ecology investigated here will be translated into a public display seen by hundreds of children and members of the public. The PIs will engage a K-12 teacher in the fieldwork at sea through a "Science Teachers Aboard Research Ships (STARS)" program and will recruit an undergraduate researcher through the CMORE Scholars program at the University of Hawaii.

The advantages and drawbacks of a mixotrophic strategy will depend on the availability of resources and competitors and the likelihood of viral infection. The timing of grazing will be tested to determine whether Florenciella grazes continuously or separates it grazing and photosynthetic activities by only feeding at night. Prey preferences of Florenciella will be tested in competitive grazing experiments offering Prochlorococcus as prey in the presence of varying amounts of other bacteria and cyanobacteria. Electron microscopy will be used to determine whether prey and virus enter Florenciella by the same pathway and whether the presence of prey competitively interferes with viral infection. The kinetics of grazing by Florenciella and infection of Florenciella by FloV1 will be quantified. The results from these lab experiments will be used to parameterize a numerical model. The model will be used to answer questions and make predictions about the dynamics of the mixotroph-virus-prey system and those predictions will be compared to field data. Collectivity, these observational, experimental and quantitative analyses will provide a detailed exploration of the ecological complexity hidden at the base of the marine food web.

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#### Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	<u>OCE-1559356</u>

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