Copepod Pressure Vessel Experiments from samples collected from the East Pacific Rise during ROV SuBastian deployments and R/V Falkor (too) cruise FKt230627 in June-July 2023

Website: https://www.bco-dmo.org/dataset/923000
Data Type: experimental
Version: 1
Version Date: 2024-03-20

Project
» The Underworld of Hydrothermal Vents (Vent Underworld)

Contributors	Affiliation	Role
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Abstract
These data show results from LT50 experiments from deep-sea hydrothermal vent copepods (family Dirivultidae) living associated with pompeii worm and mussel/tubeworm habitats at the 9°50’ North East Pacific Rise. Specimens were collected with a ROV at the vent sites Tica, Biovent and Bio9 in ~2500 meters water depth during expedition FKt230627 in July 2023. Experiments were performed onboard the research vessel R/V Falkor(too). Copepods were transferred to glass vials which were then placed into pressure vessels and pressurized. Diverse exposure times, temperatures, pressures, and oxic/anoxic conditions were selected to explore the tolerance of deep-sea hydrothermal vent copepods in relation to habitat, temperature, oxygen concentration and pressure. The data were collected by Alessandro Messora, under the supervision of Dr. Sabine Gollner (Royal Netherlands Institute for Sea Research (NIOZ), NL) and Prof. Monika Bright (University of Vienna, AUT).

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Coverage

Location: Near 9 North on EPR at vent sites Tica, Biovent, and Bio9
Spatial Extent: **N**:9.98494 **E**:-104.29138 **S**:9.83846 **W**:-104.294
Temporal Extent: 2023-07-07 - 2023-07-21

Dataset Description

Methods & Sampling

Specimens were collected with ROV SuBastian at 9°N 50'N and 104°W along the East Pacific Rise in the Tica vent area and sites Bio9 and Biovent (~2500 m depth) during expedition FKt230627 in July 2023 aboard R/V Falkor(too).

Deep-sea hydrothermal vent copepods were sampled from two different habitats: diffuse flow habitats dominated by *Riftia pachyptila* tubeworms and *Bathymodiolus thermophilus* mussels, and focused flow habitats dominated by *Alvinella pompejana* pompeii worms. A suction sampler and/or ROV manipulator grabs were used to sample detritus and megafauna respectively. These samples were then sorted for copepods and subsampled to be used for a total of 118 LT50/LD50 exposure incubations performed on board the research vessel. Each incubation contained 3 replicate vials with 10 copepods each, for a total of 3540 copepods.

Diverse exposure times, temperatures, pressures, and oxic/anoxic conditions were selected to explore the tolerance of deep-sea hydrothermal vent copepods in relation to habitat, temperature, oxygen concentration and pressure. Exposure times were the following: 2h, 4h, 8h, 10h. Temperature ranges for each incubation were established based on the ROV's temperature probe readings. A control temperature of 4°C was always included to represent similar thermal conditions to the ambient bottom water around hydrothermal vents. The other temperatures (12, 22, 25, 28, 31, 34, 37, 40, 43°C) were selected to obtain a complete survival curve that ranged from ~100% survival at the lowest temperature to ~0% at the highest temperature for each habitat/time/oxygen/pressure treatment combination. Two oxygen concentration conditions were tested: oxic (163.88 ± 5.82 µM before and 95.63 ± 9.14 µM after incubations) and anoxic (0.24 ± 0.11 µM before and 1.76 ± 0.38 µM after incubations). Concentrations were monitored before and after incubations using self-adhesive Oxygen Sensor Spots mounted on the inside of the incubation vials (SP-PSt3-SA) and a fiber-optic oxygen meter (Fibox 4).

Ten live copepods were transferred using a pipette to each seawater-filled 5.9 ml glass vial (Exetainer 719W) equipped with an airtight pressure relief cap. The glass vials were placed into pressure vessels and pressurized up to 200 bar (3000 psi) using a Waters 515 HPLC Pump, and then placed in a water bath (ARGOLAB WB 12 Lt) to keep a constant temperature. At the end of each incubation the vessel was depressurized, and the content of each vial was examined under a Leica EZ4 W stereomicroscope to record the number of living/dead copepods. After the experiment, dead copepod specimens were not disposed, but instead were fixed in 95% Ethanol for preservation.

The data was used to estimate the median lethal temperature (LD50) corresponding to each exposure time (LT50) by fitting a survival curve to the temperature and respective copepod survival data using Nonlinear Least Squares regression and isolating the theoretical 50% survival temperature.

Data Processing Description

At the end of each incubation the vessel was depressurized, and the content of each vial was examined under a stereomicroscope to record the number of living/dead copepods. Data include the counts of living/dead copepods per experiment. The data was used to estimate the median lethal temperature (LD50) corresponding to each exposure time (LT50) by fitting a survival curve to the temperature and respective copepod survival data using Nonlinear Least Squares regression and isolating the theoretical 50% survival temperature.

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Related Publications

Klose, J., Polz, M. F., Wagner, M., Schimak, M. P., Gollner, S., & Bright, M. (2015). Endosymbionts escape dead hydrothermal vent tubeworms to enrich the free-living population. Proceedings of the National Academy of Sciences, 112(36), 11300–11305. https://doi.org/10.1073/pnas.1501160112
Methods

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Parameters

Parameters for this dataset have not yet been identified

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Instruments

Dataset-specific Instrument Name	ARGOLAB WB 12 Lt
Generic Instrument Name	ARGOLAB Water Bath 12 Liters
Dataset-specific Description	Glass vials were placed into pressure vessels and placed in a water bath (ARGOLAB WB 12 Lt) to keep a constant temperature.
Generic Instrument Description	ARGOLab waterbath without recirculation pump. Working range: +5°C above ambient temperature to 100°C. Tank capacity: 12 liters. Electronic temperature regulator with integrated PID control Visual and acoustic alarms that can be silenced by the operator. Safety class 2 according to DIN 12880. Temperature stability at 37 °C: ± 0.1 °C. Temperature accuracy: ± 0.2 °C. Digital timer with programming range 1 min - 99 h and 59 min, and continuous operation. Rack capacity: up to 4 modules. Tank dimensions (W x H x D): 300 x 240 x 200 mm Weight: 12 Kg. Power supply: 220 ± 10% V / 50-60 Hz, Power: 900 W.

Dataset-specific Instrument Name	Leica EZ4 W Compound Microscope
Generic Instrument Name	Microscope - Optical
Dataset-specific Description	At the end of each incubation the content of each vial was examined under a stereomicroscope to record the number of living/dead copepods.
Generic Instrument Description	Instruments that generate enlarged images of samples using the phenomena of reflection and absorption of visible light. Includes conventional and inverted instruments. Also called a "light microscope".

Dataset-specific Instrument Name	ROV SuBastian Multi-Chamber Suction Sampler
Generic Instrument Name	Multi-chamber Suction Sampler
Dataset-specific Description	Specimens were collected with ROV SuBastian suction sampler at the vent sites.
Generic Instrument Description	An underwater device that collects animals and other samples under gentle suction and deposits them into a collection chamber. Also known as an oceanographic 'slurp' sampler. The primary components are an electrical or hydraulic pump, outlet hose connected to collection chamber, and inlet hose.

Dataset-specific Instrument Name	Fibox 4 stand-alone fiber optic oxygen meter
Generic Instrument Name	Oxygen Sensor
Dataset-specific Description	Oxygen levels were measured with a Fibox 4 fiber optic oxygen meter to monitor whether the samples were oxic or anoxic.
Generic Instrument Description	An electronic device that measures the proportion of oxygen (O2) in the gas or liquid being analyzed

Dataset-specific Instrument Name	Self-adhesive Oxygen Sensor Spot SP-PSt3-SA
Generic Instrument Name	Oxygen Sensor Spot
Dataset-specific Description	(*** Need informaiton from the submitter to be put in here about how they were used...***)
Generic Instrument Description	Oxygen sensor spots allow oxygen measurements in closed sample containers with contactless read-out through a transparent window (glass, acryl glass) with special adapters and optical fibers. These patches often have a self-adhesive option.

Dataset-specific Instrument Name	customized pressure vessel
Generic Instrument Name	Pressure Vessel
Dataset-specific Description	Pressure vessels are custom items (see Klose et al. 2015; www.pnas.org/cgi/doi/10.1073/pnas.1501160112)
Generic Instrument Description	A pressure vessel is a container designed to hold gases or liquids at a pressure substantially different from the ambient pressure. Construction methods and materials may be chosen to suit the pressure application, and will depend on the size of the vessel, the contents, working pressure, mass constraints, and the number of items required. Examples include glassware, autoclaves, compressed gas cylinders, compressors (including refrigeration), vacuum chambers and custom designed laboratory vessels.

Dataset-specific Instrument Name	Waters 515 HPLC Pump
Generic Instrument Name	Pump
Dataset-specific Description	Glass vials were placed into pressure vessels and pressurized using a Waters 515 HPLC Pump up to 200 bar (3000 psi).
Generic Instrument Description	A pump is a device that moves fluids (liquids or gases), or sometimes slurries, by mechanical action. Pumps can be classified into three major groups according to the method they use to move the fluid: direct lift, displacement, and gravity pumps

Dataset-specific Instrument Name	ROV SuBastian
Generic Instrument Name	ROV SuBastian
Dataset-specific Description	Specimens were collected with ROV SuBastian at the vent sites Tica, Biovent and Bio9 using suction sampler and/or manipulator grabs.
Generic Instrument Description	ROV SuBastian is operated from the research vessel Falkor and the R/V Falkor(too). The ROV is outfitted with a suite of sensors and scientific equipment to support scientific data and sample collection, as well as interactive research, experimentation, and technology development. More information available at https://schmidtocean.org/technology/robotic-platforms/4500-m-remotely-op...

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Deployments

FKt230629

Website	https://www.bco-dmo.org/deployment/923061
Platform	R/V Falkor (too)
Report	https://service.rvdata.us/data/cruise/FKt230629/doc/FKt230629_expedition_report.pdf
Start Date	2023-06-29
End Date	2023-07-28
Description	R/V Falkor (too) cruise to the East Pacific Rise (9°N 104°W) as part of the project "The Underworld of Hydrothermal Vents" which used the sub ROV SuBastian

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Project Information

The Underworld of Hydrothermal Vents (Vent Underworld)

Website: https://schmidtocean.org/cruise/underworld-of-hydrothermal-vents/

Coverage: East Pacific Rise

Since the discovery of deep-sea hydrothermal vents in 1977, scientists realize that life exists above and below the seafloor. The extent to which they are interconnected, however, remains poorly understood. We propose to characterize subsurface biosphere diversity, from viruses to animals, at deep-sea hydrothermal vents to elucidate the nature and extent of connectivity between the surficial and subsurface biospheres. We will test the hypotheses that 1) eukaryotic life is also an integral component of the subseafloor biosphere; 2) the subseafloor is inhabited by both cosmopolitan and endemic protists and fungi; and 3) the subseafloor habitats harbor larvae -and perhaps adult life stages- of vent endemic animals. This research will transform our understanding of the ecology/evolution of subseafloor habitats and our thinking about animal recruitment at deep-sea vents. Consistent with SOI's mission, we will expand our knowledge of limits of eukaryotic life. During a three-week cruise with R/V Falkor and ROV SuBastian to "East Pacific Rise" vents at the 9º50'N region, we will use in situ and lab experiments, molecular identification with amplicon sequencing, Illumina NextSeq 500 technology, 2bRAD population genetic analyses, and RT qPCR for functional analyses. Our data management plan includes deposition of metadata/samples to long-term repositories. Outreach includes inquiry-learning

telepresence programs for children from USA and Europe.

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Funding

Funding Source	Award
Schmidt Ocean Institute (SOI)	FKt230627
Netherlands Organization of Scientific Research (NWO, The Hague, The Netherlands)	OCENW.M.22.080 (Subsurface life at deep-sea hydrothermal vents)

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