ZooSCAN images of zooplankton collected along physical gradients during OAPS MOCNESS tows during R/V Oceanus cruise OC473 in the northwest Atlantic in 2011 and R/V New Horizon cruise NH1208 in the northeast Pacific in 2012 and imaged in 2021-2022

Website: https://www.bco-dmo.org/dataset/865757 Data Type: Cruise Results Version: 1 Version Date: 2024-04-15

Project

» Quantifying the drivers of midwater zooplankton community structure (Zooplankton Gradients)

Contributors	Affiliation	Role
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Abstract

This dataset consists of the imaging portion of the study described below and includes ZooScan images of zooplankton collected during OAPS MOCNESS tows during R/V Oceanus cruise OC473 in the Northwestern Atlantic in 2011 and R/V New Horizon cruise NH1208 in the Northeastern Pacific in 2012. Day and night stations were sampled between 0 to 1000m depths from 35 to 50 N in the northwest Atlantic in 2011, and from 35 and 50N along CLIVAR line P17N in 2012. These representative subsamples of the formalin-preserved zooplankton community from each net were imaged in 2021 and 2022. Project description: The objective of this study was to determine how environmental variables shape zooplankton community structure in the midwater. Our primary overarching hypothesis was that the abundance and size class distribution of the zooplankton community are decoupled and are influenced by different environmental variables. Furthermore, differences in zooplankton community composition and diversity in the observed distinct oceanic biogeographical provinces additionally influences both factors. Since zooplankton contributions to biogeochemistry are size dependent, standard descriptions of zooplankton community (biomass, which is a product of size and abundance) are insufficient to generate a predictive understanding of the role of zooplankton in biogeochemical cycles. The project uses particle imaging technology and metabarcoding of archived biological samples in conjunction with open access hydrographic data from two cruises conducted in the N. Atlantic and N. Pacific to test these hypotheses.

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Dataset Description

Acknowledgment:

This dataset utilized samples and data funded by a previous NSF award (OCE-1041068) "Horizontal and Vertical Distribution of Thecosome Pteropods in Relation to Carbonate Chemistry in the Northwest Atlantic and Northeast Pacific (OAPS)" <u>https://www.bco-dmo.org/project/2154</u>.

Methods & Sampling

Location:

Northwestern Atlantic, 0 to 1000m depth, 35N, 52W to 50N, 42W along CLIVAR/WOCE line A20 Northeastern Pacific, 0 to 1000m depth, 50N, 150W to 35N, 135W along CLIVAR/WOCE line P17N

Methodology:

To obtain samples, a 1m2 Multiple Opening/Closing Net and Environmental Sensing System (MOCNESS; Wiebe et al., 1985) equipped with nine 150 m nets was deployed during the mid-day and mid-night on cruises carried out as described in dataset "MOC aqui log sheets" (<u>https://www.bco-dmo.org/dataset/3546</u>). Briefly, sampling was at consistent intervals including 1000-800, 800-600, 600-400, 400-200, 200-100, 100-50, 50-25, 25-0m at day/night stations from 35 to 50 N in the northwest Atlantic in 2011, and from 35 and 50N along CLIVAR line P17N in 2012.

Upon retrieval, the catch from each of the eight discrete nets were divided into splits. One-half of a sample was preserved in 95% ethanol, one quarter was preserved in 5% buffered formalin, and one quarter was used for live viewing and picking, and then preserved in 70% ethanol.

A representative subsample of the formalin-preserved zooplankton community from each net were imaged using a ZooSCAN ver. 4 at 4,800 dpi (following the methods in: Gorsky et al., 2010, Vandromme et al., 2012, as detailed in Maas et al. 2021). In order to better represent all size classes in the images, the original sample was divided into three size categories. All individuals larger than 2 cm were selected by eye and scanned separately from all the others ("d1"). The remainder of the sample was sieved through a 1-mm mesh sieve, and both size fractions ("d2" >1000um but excluding d1; "d3" between 150-1000um) were individually scanned. From these smaller size fractions, at least 1500 particles were scanned after subsampling using a Motoda splitter (Motoda, 1959), requiring generation of two separate scans for both size classes. This resulted in a total of five images per net.

Image names

Image names include: cruise#_mocnessID_net#_sizefraction_ and _a|b if a replicate and end in _1.tif

Multiple images of the same size fraction were sometimes taken to obtain a sufficient number of particles. These replicates are named a or b. If there is no replicate they don't have a letter in the image name. An a and b scan were always done for size classes d2 and d3. This was important because the split size is for the sum of a+b (e.g. if a is $\frac{1}{4}$ and b is $\frac{1}{4}$, the acq_sub_part will be 0.5).

Example of image names:

ae1830_m13_n4_d3_a_1.tif [a replicate] ae1830_m13_n4_d3_b_1.tif [a replicate] ae1830_m13_n5_d1_1.tif [no replicate]

Related Datasets may contain the "object_id" (the particle/organism id) which is constructed the same way as the image name except it as an additional _# at the end. This additional number in the object_id is added by the Zooprocess software (Hydroptic, 2016).

e.g. object_id: ae1614_m3_n1_d2_a_1**_100** image_name: ae1614_m3_n1_d2_a_1.tif

Instruments summaries:

MOCNESS

The Multiple Opening/Closing Net and Environmental Sensing System or MOCNESS is a family of net systems based on the Tucker Trawl principle. There are currently 8 different sizes of MOCNESS in existence which are designed for capture of different size ranges of zooplankton and micro-nekton. Each system is designated according to the size of the net mouth opening and in two cases, the number of nets it carries. The original MOCNESS (Wiebe et al, 1976) was a redesigned and improved version of a system described by Frost and McCrone (1974)(from MOCNESS manual). These nets allow for the discrete sampling of zooplankton at targeted depths while simultaneously capturing data on physical parameters. The MOCNESS used in all sampling for both cruises was a 1m2 (mouth size) rigged as below.

From OC473 Cruise Report (Lawson, et al. (2011), http://hdl.handle.net/1834/43091):

"The MOCNESS was equipped with eight 150-um mesh nets (nets 1-8; borrowed from URI) and one 333-um mesh net (net 0). The underwater unit used was #169. In addition to the standard temperature and conductivity probes the system also had a beta-type strobe-light unit for reducing avoidance of the nets by some zooplankton and possibly small fish. The strobe system has two units each with 12 LED sets (LUXEON Rebel LED) with peak output between 490-520 nm. Seven of the 24 LED sets were no longer working at the start of the sampling. The LEDs are powered by the MOCNESS battery and their pulse width, amplitude, flash rate period, and on/off are controlled by the MOCNESS software. For this cruise the pulse width was 2 ms, the relative amplitude was 99%, and the flash interval was 100 ms."

From NH1208 Cruise Report (Lawson, et al. (2012), http://hdl.handle.net/1834/43090)

"The MOCNESS was equipped with eight 150-um mesh nets (nets 1-8; borrowed from URI) and one 333-um mesh net (net 0). The system was equipped with the standard SeaBird temperature and conductivity probes (units #535 and #120 respectively). The underwater unit used was #169. In addition to the standard temperature and conductivity probes the system also had a beta-type strobe-light unit for reducing avoidance of the nets by some zooplankton and possibly small fish. The strobe system has two units each with 12 LED sets (LUXEON Rebel LED) with peak output between 490-520 nm. Seven of the 24 LED sets were no longer working at the start of the sampling. The LEDs are powered by the MOCNESS battery and their pulse width, amplitude, flash rate period, and on/off are controlled by the MOCNESS software. For this cruise the pulse width was 2 ms, the relative amplitude was 99%, and the flash interval was 100 ms. The strobe unit was only used for the first four tows, after which problems with blowing the underwater unit 5A fuse (a symptom typical of strobe unit problems from the past) led us to disconnect it and not use it for the remainder of the tows."

ZOOSCAN

The ZooSCAN (CNRS patent) system makes use of scanner technology with custom lighting and a watertight scanning chamber into which liquid zooplankton samples can be placed. The scanner recovers a high-resolution, digital image and the sample can be recovered without damage. These digital images can then be investigated by computer processing. While the resolution of the digitized zooplankton images is lower than the image obtained using a binocular microscope, this technique has proven to be more than adequate for large sample sets. Identification of species is done by automatic comparison of the image (vignette) of each individual animal in the scanned image with a library data set which may be built by the investigator for each individual survey or imported from a previous survey. The latest machine learning algorithm allows high recognition levels even if we recommend complementary manual sorting to achieve a high number of taxonomic groups. Scans for this dataset performed with a ZooScan (Hydroptic, HYDROPTIC_V4) running with Vuescan (version 9.5.24) and Zooprocess (version 8.22, ImageJ macro suite). Images were taken at either 4800dpi with a narrow frame or 2400dpi with a large frame.

Data Processing Description

Scans were processed using Zooprocess (version 8.22, ImageJ macro suite). The "Convert and process from RAW" function was used to separate particles into individual vignettes and generate a suite of measurements for each particle. "Doubles" (vignettes containing more than one particle) were manually separated in the software and reprocessed.

BCO-DMO Processing Description

BCO-DMO Data Manager Processing Notes (Version 1):

* Images bundled for download in the Data Files section in .zip bundles by mocness tow.

* The filesize and md5 checksum was added to the image metadata table.

** After images were transferred to BCO-DMO, a file inventory was made containing the filename, filesize in bytes, and checksum (md5sum). This file inventory table was joined with the provided metadata table to verify the file collection is complete without any missing files.

Problem Description

Some chaetognaths and all pteropods were removed prior to imaging in association with the original OAPS and ancillary projects.

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Related Publications

Gorsky, G., Ohman, M. D., Picheral, M., Gasparini, S., Stemmann, L., Romagnan, J.-B., ... Prejger, F. (2010). Digital zooplankton image analysis using the ZooScan integrated system. Journal of Plankton Research, 32(3), 285–303. doi:<u>10.1093/plankt/fbp124</u> *Methods*

Hydroptic (2016). ZooSCAN. Available at

http://www.hydroptic.com/index.php/public/Page/product_item/ZOOSCAN. Accessed June 17th, 2021. Software

Lawson, G., Wiebe, P., Wang, Z. A., Lavery, A., Maas, A. E., Blanco-Bercial, L., Hoering, K. A., Copley, N., Fincke, J., White, T. Bergan, A. J., Wurtzell, K.(2012). R/V Oceanus Cruise #473 'Ocean Acidification Pteropod Study' Cruise Report. [Aquadocs]. Retrieved from <u>http://hdl.handle.net/1834/43091</u> *Methods*

Lawson, G., Wiebe, P., Wang, Z. A., Lavery, A., Maas, A. E., Copley, N.,Blanco-Bercial, L., Hoering, K. A., Bolmer, T., Chu, S.,Roger, L., Crockford, T. (2012). R/V New Horizon Cruise #1208 'Ocean Acidification Pteropod Study' Cruise Report. Retrieved from <u>http://hdl.handle.net/1834/43090</u> *Methods*

Maas, A. E., Gossner, H., Smith, M. J., & Blanco-Bercial, L. (2021). Use of optical imaging datasets to assess biogeochemical contributions of the mesozooplankton. Journal of Plankton Research, 43(3), 475–491. doi:<u>10.1093/plankt/fbab037</u> *Methods*

Motoda, S. (1959) Devices of simple plankton apparatus. Memoirs of the Faculty of Fisheries Hokkaido University, 7, 73-94. Available from http://hdl.handle.net/<u>2115/21829</u>. *Methods*

Picheral M, Colin S, Irisson J-O (2017). EcoTaxa, a tool for the taxonomic classification of images. http://ecotaxa.obs-vlfr.fr Methods

Schneider C A Pashand

Schneider, C. A., Rasband, W. S., ... (n.d.). ImageJ. US National Institutes of Health, Bethesda, MD, USA. Available from <u>https://imagej.nih.gov/ij/</u> Software

Vandromme, P., Stemmann, L., Garcia-Comas, C., Berline, L., Sun, X., & Gorsky, G. (2012). Assessing biases in computing size spectra of automatically classified zooplankton from imaging systems: A case study with the ZooScan integrated system. Methods in Oceanography, 1-2, 3–21. doi:<u>10.1016/j.mio.2012.06.001</u> *Methods*

Wiebe, P. H., Morton, A. W., Bradley, A. M., Backus, R. H., Craddock, J. E., Barber, V., ... Flierl, G. R. (1985). New development in the MOCNESS, an apparatus for sampling zooplankton and micronekton. Marine Biology, 87(3), 313–323. doi:10.1007/bf00397811 <u>https://doi.org/10.1007/BF00397811</u> [table of contents | back to top]

Related Datasets

References

Lawson, G., Wiebe, P. H. (2011) **MOCNESS logs from R/V Oceanus, R/V New Horizon OC473, NH1208** in the western North Atlantic, transect between 35 and 50N along CLIVAR line P17N from 2011-2012 (OAPS project). Biological and Chemical Oceanography Data Management Office (BCO-DMO). Version Date 2011-09-27 http://lod.bco-dmo.org/id/dataset/3546 [view at BCO-DMO] *Relationship Description: Original MOCNESS logs associated with scanned samples (images).*

IsRelatedTo

Lawson, G., Wiebe, P. H. (2012) **CTD data collected from MOCNESS hauls on R/V Oceanus and R/V New Horizon in the western North Atlantic from 2011-2012 (OAPS project).** Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 20120208) Version Date 2012-02-08 http://lod.bco-dmo.org/id/dataset/3569 [view at BCO-DMO] *Relationship Description: CTD Data associated with original MOCNESS tows (Original MOCNESS logs associated with scanned samples (ZooSCAN images)).*

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Parameters

Parameters for this dataset have not yet been identified

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Instruments

Dataset- specific Instrument Name	1m MOCNESS
Generic Instrument Name	MOCNESS
Generic Instrument Description	land in two cases, the number of nets it carries. The original MOV NESS (Muche et al. 1976) was al

Dataset- specific Instrument Name	ZooSCAN ver. 3
Generic Instrument Name	ZooSCAN
Dataset- specific Description	http://www.hydroptic.com/index.php/public/Page/product_item/ZOOSCAN
	Description excerpt from Hydroptic website http://www.hydroptic.com/index.php/public/Page/product_item/ZOOSCAN The ZooSCAN (CNRS patent) system makes use of scanner technology with custom lighting and a watertight scanning chamber into which liquid zooplankton samples can be placed. The scanner recovers a high-resolution, digitial image and the sample can be recovered without damage. These digital images can then be investigated by computer processing. While the resolution of the digitized zooplankton images is lower than the image obtained using a binocular microscope this technique has proved to be more than adequate for large sample sets. Identification of species is done by automatic comparison of the image (vignette) of each individual animal in the scanned image with a library data set which may be built by the investigator for each individual survey or imported from a previous survey. The latest machine learning algorithm allows high recognition levels even if we recommend complementary manual sorting to achieve a high number of taxonomic groups.

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Deployments

OC473

0C473	JC473		
Website	https://www.bco-dmo.org/deployment/58720		
Platform	R/V Oceanus		
Report	http://hdl.handle.net/1834/43091		
Start Date	2011-08-07		
End Date	2011-09-01		
Description	The primary objective of the proposed research is to quantify the distribution, abundance, species composition, shell condition, and vertical migratory behavior of oceanic thecosome pteropods in the northwest Atlantic and northeast Pacific, and correlate these quantities to hydrography and concurrent measurements of carbonate chemistry, including vertical and horizontal distributions of aragonite saturation. During OC473, the first cruise in the Atlantic, a combination of underway data collection and station activities will be conducted along a transect spanning 15 degrees of latitude (35° to 50° N) in the northwest Atlantic, employing six instrument packages: (1) a 1-m2 MOCNESS plankton net system; (2) a profiling Video Plankton Recorder / CTD package, including bottles for water sampling; (3) a deep (500m) towed broadband acoustic scattering system ; 94) a hull-mounted narrowband multi-frequency acoustic scattering system. It is possible that the hull mounted transducers will suffer from noise when the vessel is underway and so as a backup we will have a surface-towed sled with a backup complement of transducers; 5) an underway multi-parameter inorganic carbon analyzer and 6) a suite of chemistry-related instruments including a DIC auto-analyzer for discret bottle sample analysis, an alkalinity auto-titrator for bottle analysis and an Agilent spectrophotometer for discrete pH measurement. Supporting documentation:Cruise track image Cruise information and original data are available from the NSF R2R data catalog.		

Website	https://www.bco-dmo.org/deployment/58830	
Platform	R/V New Horizon	
Report	http://hdl.handle.net/1834/43090	
Start Date	2012-08-09	
End Date	2012-09-18	
Description	The primary objective of this cruise was to quantify the distribution, abundance, species composition, shell condition, and vertical migratory behavior of oceanic thecosome pteropods in the northeast Pacific, and correlate these quantities to concurrent measurements of carbonate chemistry. Underway data collection and station activities were conducted on a transect running between 35 and 50N along CLIVAR line P17N. Six instrument types were used: (1) a 1-m2 MOCNESS plankton net system and a 1-m diameter Reeve net; (2) a profiling Video Plankton Recorder mounted on the CTD package that includes a Rosette system with Niskin bottles for water sampling; (3) a deep (500 meter) towed broadband acoustic scattering system; (4) a surface narrowband multi-frequency acoustic scattering system; (5) an underway multi-parameter inorganic carbon analyzer and a GO underway pCO2 system; and (6) a suite of chemistry-related lab instruments for bottle sample analysis including a DIC autoanalyzer, an alkalinity auto-titrator, and an Agilent spectrophotometer for pH measurement. The R/V New Horizon departed from Newport OR, and set a course for the transect start point at 50N 150W. Following instrument package test deployments over the continental shelf, the transect ran in a single zig-zag between the start point and the end at 35N 135W; a total of 34 stations were sampled along the transect, every 1/2 degree of latitude. In addition 10 other stations were sampled with a Reeve net for live experimental pteropods. The science party, divided into biology and chemistry teams conducted 24-hour operations. Cruise information and original data are available from the NSF R2R data catalog.	

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Project Information

Quantifying the drivers of midwater zooplankton community structure (Zooplankton Gradients)

Website: https://www.nsf.gov/awardsearch/showAward?AWD_ID=1948162

Coverage: North Atlantic and North Pacific

NSF Award Abstract:

Processes in the midwater region below 200 m depth, also known as the twilight zone, represent a major unknown for the biology and chemistry of the ocean. Studies of animals drifting in the oceans, known as zooplankton, are scarce due to the difficulty and associated time and costs of sampling deep waters. The advent of automated image analysis and genetic tools is leading to a rapid increase in our knowledge of the diversity, abundances and size distribution of communities in shallow waters. However, our understanding of the deeper layers of the ocean is still in its infancy, and there are few studies that combine these three facets of the ecology of the zooplankton. The objective of this project is to leverage existing samples, obtained from previously NSF-funded research in the North Pacific and North Atlantic, to study how the abundances, diversity, and size distribution of zooplankton in the midwater vary with latitude and environmental factors. Automated image analyses provide information on abundance and size, and genetic analyses give unprecedented data on the diversity of the midwater community for the North Atlantic and the North Pacific, from subtropical to subarctic environments. This project provides high quality hands-on training opportunities for at least two undergraduate researchers and generates material for undergraduate and graduate courses. Two workshops train educators on the classroom use of the NSF-funded Biological and Chemical Oceanography Data Management Office (BCO-DMO) open access oceanographic data.

Recent advances in image analysis and metabarcoding of zooplankton communities via new data tools are an opportunity to generate quantitative and predictive relationships between environmental drivers and

zooplankton diversity, abundances and size distribution. While this information is available for plankton in epipelagic regions, the focus of this study is on midwater communities, which remain poorly characterized. Obtaining these data is the first step towards a quantitative analysis that assesses the impact of the midwater community on biogeochemical cycles. The project uses archived samples from two cruises conducted in the N. Atlantic and N. Pacific to test hypotheses about how temperature, midwater hypoxia, primary productivity and biogeographic province shape the size class structure, biodiversity and behavior (diurnal vertical migration) of zooplankton communities. These newly-generated image and metabarcoding datasets of the mesozooplankton community from 0-1000 m are cross-comparable with other ocean regions. These data describe how migratory and midwater resident zooplankton communities are structured by environmental variables and demonstrate how this influences their biogeochemical contributions (specifically active flux and midwater attenuation of flux). Data tools generated for the image analysis in combination with metabarcoding has broad application in plankton ecology and allows metanalysis of other datasets. The project is complementary to ongoing national and international projects that seek to describe the function and structure of the midwater. In contrast to existing modeling and process projects, this project covers a moderately large geographic area and thus provides a strong comparative foundation for broader community-wide assessment of the function of zooplankton in the twilight zone.

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.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1948162

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