Dataset name: **Metagenomics and exoproteomics from deep water**

|  |  |
| --- | --- |
| Parameters: | * **Genes and proteins of Bacteria and Archaea** |

PROJECT TITLE: **MOBYDICK**

Oceanographic cruise: **MOBYDICK**

Start date: **18/02/2018**

End date: **27/03/2018**

Project manager: **Bernard Quéguiner** [bernard.queguiner@mio.osupytheas.fr](mailto:bernard.queguiner@mio.osupytheas.fr)

Address: **Mediterranean Institute of Oceanolography**

**Institut Pytheas - Observatoire des Sciences de l'Univers**

**Bâtiment OCEANOMED, Campus de Luminy, case 901**

**F-13288 Marseille Cedex 09, France**

Chief scientist: **Ingrid Obernosterer** [ingrid.obernosterer@obs-banyuls.fr](mailto:ingrid.obernosterer@obs-banyuls.fr)

Address: **Laboratoire d’Océanographie Microbienne**

**Observatoire Océanologique de Banyuls sur mer**

**66650 Banyuls sur mer, France**

Geographic information: **Indian sector of the Southern Ocean**

Latitude: **49.5°S – 52.5°S**

Longitude: **67,0°E – 74.5°E**

Parameter supervisor: **Thomas Reinthaler**

Microbial Oceanography Laboratory,

University of Vienna,

Althanstrasse 14,

Vienna, Austria

+43 1 4277 764 32

[thomas.reinthaler@univie.ac.at](mailto:thomas.reinthaler@univie.ac.at)

Dataset contact: **Thomas Reinthaler**

Microbial Oceanography Laboratory,

University of Vienna,

Althanstrasse 14,

Vienna, Austria

+43 1 4277 764 32

[thomas.reinthaler@univie.ac.at](mailto:thomas.reinthaler@univie.ac.at)

# OPERATIONS

## Sampling device(s)

Water samples were collected from the rosette bottles and/or *in situ* pumps (ISP\_ISMI casts) at selected stations.

## List of stations sampled

**Table 1 : details of sampled stations**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Station ID** | **Date** | **Cast ID** | **Sampling depth** | **Bottle #** | **Pump #** | **Volume filtered** |
| M2\_1 | 26/02/18 | CTD\_02 | 450 m | 1-12 |  | 144 L |
| M2\_1 | 27/02/18 | ISP\_ISMI\_1 | 420 m |  | Pump1 | 121 L |
| M4\_1 | 02/03/18 | CTD\_18 | 4000 m | 1-18 |  | 216 L |
| M4\_1 | 03/03/18 | ISP\_ISMI\_2 | 2908 m |  | Pump1 | 268 L |
| M4\_1 | 03/03/18 | ISP\_ISMI\_2 | 1257 m |  | Pump2 | 339 L |
| M3 | 04/03/18 | CTD\_19 | 1500 m | 1-22 |  | 258 L |
| M3 | 05/03/18 | ISP\_ISMI\_3 | 1000 m |  | Pump1 | 217 L |
| M3 | 05/03/18 | ISP\_ISMI\_3 | 500 m |  | Pump2 | 296 L |
| M2\_2 | 07/03/18 | CTD\_31 | 400 m | 1-22 |  | 264 L |
| M2\_2 | 08/03/18 | ISP\_ISMI\_4 | 490 m |  | Pump1 | 223 L |
| M4\_2 | 13/03/18 | CTD\_47 | 3500 m | 5-21 |  | 192 L |
| M4\_2 | 14/03/18 | ISP\_ISMI\_6 | 3350 m |  | Pump1 | 180 L |
| M4\_2 | 14/03/18 | ISP\_ISMI\_6 | 2500 m |  | Pump2 | 189 L |
| M2\_3 | 17/03/18 | ISP\_ISMI\_7 | 490 m |  | Pump1 | 149 L |
| M2\_3 | 17/03/18 | ISP\_ISMI\_7 | 220 m |  | Pump2 | 176 L |
| M3\_3 | 18/03/18 | ISP\_ISMI\_8 | 1500 m |  | Pump1 | 262 L |
| M3\_3 | 18/03/18 | ISP\_ISMI\_8 | 800 m |  | Pump2 | 326 L |

# INSTRUMENTS

Instrument Type: **In Situ Pump**

Manufacturer: **McLane Research Laboratories**

Model: **WTS-6-1-142**

Instrument Features / Calibration: N/A

# DESCRIPTION of PARAMETERS

## Measurement details

Seawater samples were collected into 25 L polycarbonate carboys. Additionally 2 high volume *in situ* pumps per station were used to collect up to 300 L of seawater at *in situ* pressure and temperature.

**High volume sampling for proteomics and exoproteomics**

High volume samples were taken for later proteomics and exo-proteomics analysis. Per sampling around 264 L of seawater were transferred to a barrel within 15 min upon arrival of the rosette on deck. The raw seawater was pre–filtered over a 3.0 µm (147 mm diameter, Millipore) and a 1.0 µm (293 mm diameter, GE Water Tech) polycarbonate filter using positive pressure diaphragm pumps (Verderair Cont-EX). The pre–filtered seawater was filtered onto a 0.2-µm Durapore membrane (293 mm diameter, Millipore). Initially, the permeate was concentrated to ~1 L using an ultrafiltration system with a pore size of 5,000 Da and 0.5 m2 filtration area (Millipore Pellicon Ultrafiltration System). This volume was further concentrated to ~30 mL using a small scale 5,000 Da ultrafiltration cassette (Vivaflow Sartorius). The final concentrate was flash frozen in liquid nitrogen and stored at -80ºC. In the home lab the proteins from these samples will be extracted and analyzed using an nanoLC-MS/MS approach.

**High volume sampling for metagenomics using *in situ* pumps**

For sampling of deep water *in situ*, McLane large volume sampling *in situ* pumps were used. Generally, 2 depth layers were sampled and per sampling between 200 and 300 L were pumped over a sequence of filters of 3.0 µm, 0.8 µm and 0.22 µm pore sizes. In the home laboratory, DNA will be extracted for metagenomic analysis on an illumine platform.

## Analytical procedure

Samples for metagenomics and exoproteomics will be extracted immediately after the arrival of samples in the home lab. The metagenomic DNA will be analyzed using a commercial state of the art sequencing facility using Illumina or Oxford Nanopore sequencing. Endo- and exoproteins will be analyzed at VIMES (Vienna Metabolomics Center) of the University of Vienna using a nanoLC/MS QExactive System from ThermoFisher Scientific. The pipeline to analyze the gene and protein data is still subject to development.

## Units

N/A

## Sensor precision

N/A

## Post-cruise data analysis/treatment required

N/A

## Estimated Date of Delivery

Raw data after 8 months; first analysis comparing 2 stations after 13 months; dataset fully exploited after 30 months.

# BIBLIOGRAPHY

Bergauer K., Fernàndez-Guerra A., Garcia J.A.L., Sprenger R.R., Stepanauskas R., Pachiadaki M.G., Jensen O.N., Herndl G.J., 2018. Organic matter processing by microbial communities throughout the Atlantic water column as revealed by metaproteomics. *Proceedings of the National Academy of Sciences of the United States of America*, **115**, E400–E408. doi:10.1073/pnas.1708779115.

Armengaud J., Christie-Oleza J.A., Clair G., Malard V., Duport C., 2012. Exoproteomics: exploring the world around biological systems. *Expert Review of Proteomics*, **9**, 561–575. doi:10.1586/epr.12.52.