



R/V Yokosuka Cruise Report

YK14-12

Quantitative evaluation of three different organic carbon
sources to the abyssal ecosystems using *in situ*
experimental approaches

June 28th, 2014 – July 4th, 2014

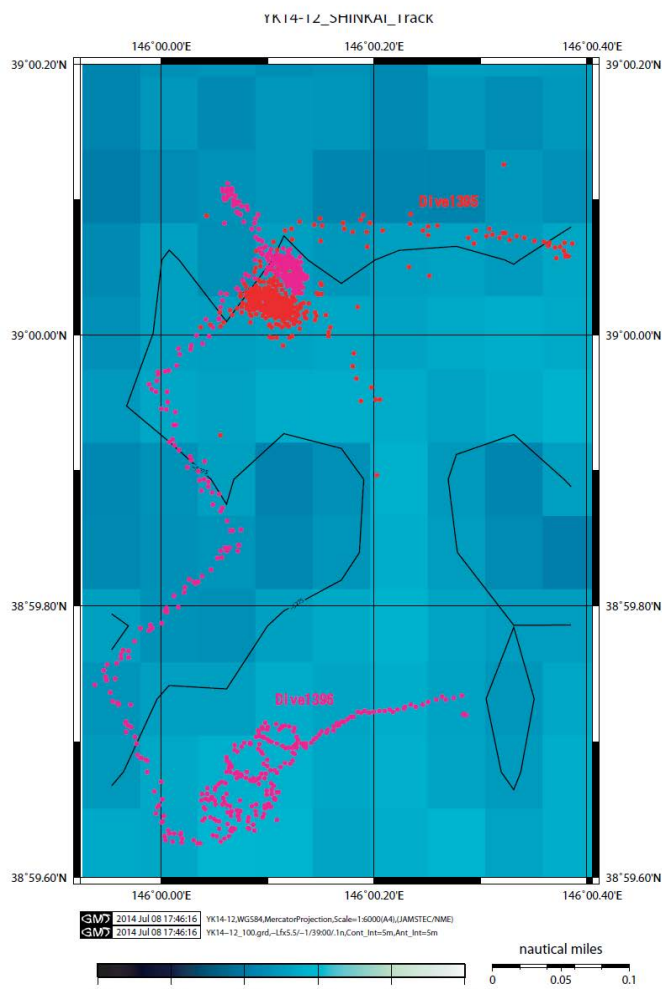
Japan Agency for Marine-Earth Science and Technology
(JAMSTEC)

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1. Cruise Information

- Cruise ID: YK14-12
- Name of vessel: R/V Yokosuka
- Title of the cruise: Quantitative evaluation of three different organic carbon sources to the abyssal ecosystems using *in situ* experimental approaches
- Title of proposal:
Quantitative evaluation of three different organic carbon sources to the abyssal ecosystems using *in situ* experimental approaches
(Hidetaka Nomaki)
- Cruise period: 28th June to 4th July, 2014
- Ports of call: Yokosuka to Yokosuka
- Research area: western North Pacific
- Research Map



2. List of participants

2.1. Onboard scientists

- Chief scientist [Affiliation]: Hidetaka Nomaki [JAMSTEC]
- Representative of the science party [Affiliation]: Hidetaka Nomaki [JAMSTEC]
- Science party (List) [Affiliation, assignment etc.]

Hidetaka NOMAKI	Japan Agency for Marine-Earth Science and Technology
Norio MIYAMOTO	Japan Agency for Marine-Earth Science and Technology
Manabu NISHIZAWA	Japan Agency for Marine-Earth Science and Technology
Miho HIRAI	Japan Agency for Marine-Earth Science and Technology
Tomo KITAHASHI	Atmosphere and Ocean Research Institute, The University of Tokyo
Yuki UEJIMA	Japan Agency for Marine-Earth Science and Technology
Ryuji KONDO	Fukui Prefectural University
Katsuaki MATSUDA	Fukui Prefectural University
Sayaka YAMAGUCHI	Fukui Prefectural University
Tadanobu MARUYAMA	Fukui University of Technology
Akira SHIHONGI	Fukui University of Technology
Teresa Amaro	Hellenic Center of marine Research

Marine Technician

Satomi Minamizawa	Nippon Marine Enterprises, LTD
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2.2. Shinkai 6500 operation team

Submersible Op. Manager	Toshiaki SAKURAI
Deputy Submersible Op. Manager	Yoshitaka SASAKI
1st Submersible Staff	Kazuki IJIMA
1st Submersible Staff	Tetsuya KOMUKU
1st Submersible Staff	Mitsuhiro UEKI
1st Submersible Staff	Keita MATSUMOTO
2nd Submersible Staff	Hirofumi UEKI
2nd Submersible Staff	Keigo SUZUKI
2nd Submersible Staff	Fumitaka SAITO
2nd Submersible Staff	Yudai TAYAMA
2nd Submersible Staff	Masaya KATAGIRI

3rd Submersible Staff

Takuma GOTO

2.3 Crew members

Captain	Yoshiyuki NAKAMURA
Chief Officer	Yasuhiko SAMMORI
2nd Officer	Takeshi EGASHIRA
3rd Officer	Tomohiro YUKAWA
Jr. 3rd Officer	Yusuke ISHI
Chief Engineer	Tadashi ABE
1st Engineer	Wataru KUROSE
2nd Engineer	Kenta IKEGUCHI
3rd Engineer	Koichi HASHIMOTO
Jr. 3rd Engineer	Kazuki ONO
Chief Electronics Operator	Hiroyasu SAITAKE
2nd Electronics Operator	Misato HATA
Boat Swain	Hatsuo ODA
Quarter Master	Yasuo KONNO
Quarter Master	Tsuyoshi CHIMOTO
Quarter Master	Hiroaki NAGAI
Sailor	Yoshihiro OGAWA
Sailor	Yuta MOTOOKA
Sailor	Kento KANDA
No.1 Oiler	Toshikazu IKEDA
Oiler	Kazuo SATO
Oiler	Keita FUNAWATARI
Assistant Oiler	Toshinori MATSUI
Assistant Oiler	Seiya WATANABE
Chief Steward	Yukio TACHIKI
Steward	Hideo FUKUMURA
Steward	Toru MURAKAMI
Steward	Yoshie HIDAKA
Steward	Yohei EBIKO

3. Observation

3.1. General purpose

Abyssal plain covers roughly half areas of the Earth Surface, thus consist largest marine ecosystem on Earth and contains abundant benthic fauna living on and in the sediment. Abyssal plain ecosystem has been thought to be sustained by POM fluxes from the photic zone, which originated from photosynthesis. Since surface productivity of the ocean differs largely with latitude, distance from land, upwelling intensity, and so on, organic matter fluxes to the seafloor also varied with oceanic settings. Those differences in POM fluxes to the seafloor are believed to be major controlling factor of benthic diversity, biomass, sediment community oxygen consumption, etc. On the other hand, recent studies on sediment metagenomics reveal that some chemolithoautotrophic microbes may inhabit even in the “normal” deep-sea floor where no active hydrothermal vent or hydrocarbon seepage exist. However, there is no report on chemoautotrophic production rate at the deep-sea floor. It also become apparent that episodic food supply to the seafloor like jellyfish fall provide significant proportion of organic matters. Those episodic food supplies should also take into account when we consider about the deep-sea biogeochemical cycles. In this study, we carried out sediment samplings and in situ incubation experiments to investigate diversity and biomass of benthos and relevant biogeochemical cycles at the deep-sea floor under the eutrophic abyssal plain.

3.2. In situ experiments to reveal autotrophic microbial activities at the abyssal plain

Hidetaka Nomaki and YK14-06, YK14-12 scientists

We carried out in situ incubation experiments together with sediment geochemistry investigation at the 39N site (39°N, 146°E, water depth = 5300m) during YK 14-6 and YK14-12 cruises. The experiment is planned to reveal the chemoautotrophic carbon production rate at the deep-sea floor using in situ incubation cores. We prepared 5 different substrates which were labeled with either ^{13}C or ^{15}N . During YK 14-12 cruise, we collected long-term incubation samples which were deployed at the YK14-06 cruise. We also deployed and recovered short-term incubation samples during YK14-12 cruise.

On board, sediment samples were sliced into different depth layers and subsampled for viral, microbial, geochemical, and meiofaunal analyses.

3.3. In situ experiments for heterotrophic organism activities

The objective of this cruise is to look at the impact of key megafauna organisms on the uptake of carbon in macro/meio/microbial fauna. For that, we performed 2 in situ experiments on the seafloor where we injected two different labelled types of food, one with diatoms and another one with cyanobacteria. The 2 in situ experiments had two different durations in time (1 days incubation and 58 days incubation).

During this cruise we retrieved the in situ experiments deployed on YK 14-06 and we deployed another situ incubation chamber at 39N during the dive #1395, which was retrieved during the following dive #1396. After this and in order to characterize the densities of the megabenthos on the seafloor, several video transects were made on the area. On board, sediment samples were sliced into different depth layers (0-1, 1-2, 2-3, 3-5, 5-10 and 10-15 cm) to look at meiofaunal and macrofauna diversity and abundances. Deep-sea holothurians were collected, dissected and preserved at -80°C freezer.

Samples taken:

*Dive 1395 –

Megabenthos

Background samples:

2 *Deima* sp.

sediment:

2 open boxes experiment for macrofauna.

3.4. Significance of nitrification in eutrophic deep-sea sediments

Takuro Nunoura, Manabu Nishizawa, Miho Hirai (JAMSTEC)

Nitrification; oxidation of ammonia and nitrite to nitrite and nitrate, respectively, is one of the most dominant chemolithotrophic metabolisms in deep sea environments, and likely play significant roles in nitrogen cycle in marine sedimentary habitats. In fact, the presence and predominance of putative ammonia-oxidizing thaumarchaeotes in benthic archaeal communities has been reported. However, little is known about their in situ activity and significance in

benthic nitrogen cycle. In this cruise, we took abyssal plain sediments from the nutrient western north Pacific site, and will examine the significance of nitrifier communities using molecular biological and stable isotopic approaches. Moreover, we conducted in situ incubation analyses to examine nitrification activity at the equatorial Pacific site. The stable isotopic pore water chemistry and molecular biological approaches to the samples obtained by the in situ incubation will provide novel insights into the benthic nitrogen cycle under the relatively oligotrophic ocean.

3.5. Virus-prokaryote interactions in abyssal ecosystems

Eugenio Rastelli and Teresa Amaro

(Department of Life and Environmental Sciences, Polytechnic University of Marche, Italy)

Introduction

Viruses are by far the most abundant biological entities in the world's oceans (approximately 4×10^{30} , Suttle 2007). Recent estimates suggest that every kg of deep-sea sediment contains 10^{12} viruses and 10^{11} prokaryotes (Sogin et al 2006). Recent studies revealed that viral infection in aquatic sediments can be the major cause of mortality for benthic prokaryotes (Danovaro et al 2008). Viral lysis transforms infected microbes into organic detritus, which can then be used again by non-infected prokaryotes and/or contribute to biogeochemical cycles. Extracellular DNA is likely to play a key role in both processes (Dell'Anno & Danovaro 2005) and can be a reservoir of genes (1 kg of deep-sea sediment can contain 10^{13} copies of 16S rRNA genes; Corinaldesi et al 2011).

Purpose

During the YK14-12 cruise organised by JAMSTEC samples were taken from the eutrophic station 39N, and the goal of the Italian research team (UNIVPM) was to investigate the dynamics in virus – prokaryotes activity and interactions in the abyssal ecosystem in the Pacific ocean and compare the results with the data acquired during the previous year's cruises in the Ogasawara trench and abyssal plain (11-2011), Japan trench (12-2012), and Mariana Trench (01-2014). Additional experiments using in situ incubation were conducted in collaboration with JAMSTEC to study the response of benthic communities to food inputs and prokaryotic chemosynthetic processes. Results from the present cruise will be compared with those obtained from the previous cruise in which the same experiments were conducted in a more oligotrophic area (station 1N, during YK13-09 cruise).

Materials and methods

Seawater samples were collected during the YK14-12 cruise in station 1N, from the sea surface and the bottom of the abyssal ecosystem in the Pacific Ocean. Moreover, surface and sub-surface sediments were collected from the same station.

Additional experiments (*in situ* temperature incubations of sediment samples) were conducted on board and *in situ*. On these samples, UNIVPM research team will conduct analyses for the determination of:

Viral abundance; total prokaryotic abundance; relative importance of Bacteria and Archaea on total prokaryotic abundance; viral production, extracellular enzymatic activities; organic matter composition and turnover; DNA and RNA analyses.

Moreover, several antibiotics were used to inhibit Archaeal or Bacterial activity in dedicated sets of samples, in order to study the effects of the host inhibition on viral production.

References

Danovaro, R. et al (2008). Nature 454: 1084-1087; Corinaldesi, C. et al (2011). Mol. Ecol. 20: 642-654. 5. Dell'Anno, A. & Danovaro, R. (2005). Science 309: 2179; Sogin, M. et al (2006). PNAS 32: 12115-12120; Suttle, C.A. (2007). Nature Rev. Microbiol. 5: 801-812;

3.6. Isolation of prokaryotic and eukaryotic microorganisms from deep-ocean

Ryuji Kondo, Sayaka Yamaguchi and Katsuaki Matsuda (Fukui Prefecture Univ)

Microbes are known to important roles in material cycles in marine environments, as well as in degradation of organic matter. Heterotrophic nanoflagellates (HNF) are significant consumers of bacteria in aquatic environments. Bacteria and eukaryotic microorganisms such as yeast act as decomposition and mineralisation of organic matter in the environments. In this cruise, HNF abundance will be investigated in the water column and the bottom sediments of the western North Pacific abyssal plain. We will also try to isolate and cultivate the prokaryotic and eukaryotic microbes in order to know their physiology.

3.7. Isolation of prokaryotic and eukaryotic microorganisms from deep-ocean

Yuichiro Kashiwayama, Akira Shihongi and Tadanori Maruyama (Fukui Univ. Technol.)

Herbivorous microbes are key organisms understanding energy flow in the pelagic marine ecosystem, playing a role connecting the planktonic microalgae as the primary producers and the higher trophic community. In pelagic oligotrophic settings, a large part of primary

production is attributed to picocyanobacteria, but their minute sizes should evade grazing by multicellular zooplanktons (i.e., filter feeders). We therefore investigate protistan grazers on such picophytoplanktons by isolating the candidates from the water column as well as detecting detoxified metabolites of chlorophylls.

3.8. Acknowledgment

We thank to the captain and crew members of R/V Yokosuka for their strong supports during the cruise. We also thank to the submersible operation manager and the Shinkai 6500 operation team for their skilful operation of the submersible.

3.9. List of observation equipments

In situ incubation core (inner diameter: 8.2cm, length: 32cm)

In situ incubation box (Surface area: 900 cm²)

Holothurian cage (Surface area: ~600 cm²)

Niskin water sampler (5L)

Van Dorn water sampler (3L)

H-type push corer (inner diameter: 8.2cm, length: 32cm)

Suction sampler with multiple canister

4. Cruise log

日付 Date	時間 Local Time	内容 Note	特記事項 Description	本船位置／気象／海象 Position/Weather/Wind/Sea condition
28-Jun-14		Sail out, proceeding to research area		06/28 12:00 (UTC+9h)
	08:00	boarded		34-51.8N, 139-57.0E
	09:00	let go all shore line, left YOKOSUKA	for N. W. Pacific Ocean	South off NOJIMASAKI
	10:00-10:30	carried out onboard education & training for scientists		Raine

	11:00-11:30	meeting with Shinkai 6500 team		ENE-5 (Fresh breeze)
	13:00	Shinkai 6500 briefing		3 (Sea slight)
				2 (Low swell long)
				Visibly: 3'
29-Jun-14		Proceed to research area		06/29 12:00 (UTC+9h)
	11:00-11:30	scientific meeting		38-05.3N、142-20.5E
				East off KINKAZAN
				Rain
				East-5(Fresh breeze)
				4 (Sea moderate)
				4 (Moderate average)
				Visibly: 2'
30-Jun-14		Carry out water sampling		06/30 12:00 (UTC+9h)
	15:00	arrived at research area	N. W. Pacific Ocean	38-05.1N、145-19.0E
	15:02	released XBT		North Pacific ocean
	15:49	started to sampling of sea water by VAN DORN water sampler	38-59.9945N, 145-59.7865E	Fine but cloudy
	17:36	finished to sampling of sea water by VAN DORN water sampler	39-00.0853N, 146-00.9992E	NNW-5 (Fresh breeze)
	18:01-18:36	carried out MBES mapping survey		4 (Sea moderate)
				4 (Moderate average)
				Visibly: 8'

01-Jul-14		Shinkai 6500 dive 1395		07/01 12:00 (UTC+10h)
	09:02	Shinkai 6500 dove and started her operation #1395		39-00.1N、146-00.3E
	09:10-09:20	scientific meeting		N. W. Pacific ocean
	11:18	landed at sea bottom	Depth = 5255m	Fine but cloudy
	15:09	left bottom	Depth = 5236m	SSW-2 (Light breeze)
	17:49	recovered Shinkai 6500 and finished the operation		2 (Sea smooth)
				2 (Low swell long)
				Visibly: 8'
02-Jul-14		Shinkai 6500 dive 1395		07/02 12:00 (UTC+10h)
	09:02	Shinkai 6500 dove and started her operation #1396		39-00.0N、146-00.1E
	11:15	landed at sea bottom	Depth = 5253m	N. W. Pacific ocean
	15:11	left bottom	Depth = 5253m	Fine but cloudy
	15:20	scientific meeting		WNW-2 (Light breeze)
	17:32	recovered Shinkai 6500 and finished the operation		1 (Sea calm)
	18:00	left research area for YOKOSUKA		2 (Low swell long)
				Visibly: 2'
03-Jul-14		proceed to YOKOSUKA		07/03 12:00 (UTC+9h)

	13:00-13:30	engine room tour		37-04.4N,142-13.8E
	14:00-16:00	Shinkai 6500 tour		East off SHIOYASAKI
				Cloudy
				SE-3 (Gentle breeze)
				2 (Sea smooth)
				1 (Low swell short)
				Visibly: 8'
04-Jul-14		Arrived at YOKOSUKA		
	10:00	disembarked at YOKOSUKA		
		finished YK14-12 cruise		

5. Notice on Using

Notice on using: Insert the following notice to users regarding the data and samples obtained.

This cruise report is a preliminary documentation as of the end of the cruise.

This report may not be corrected even if changes on contents (i.e. taxonomic classifications) may be found after its publication. This report may also be changed without notice. Data on this cruise report may be raw or unprocessed. If you are going to use or refer to the data written on this report, please ask the Chief Scientist for latest information.

Users of data or results on this cruise report are requested to submit their results to the Data Management Group of JAMSTEC.