

R/V Yokosuka Cruise Report

YK13-09

In situ experimental & sampling study to understand abyssal biodiversity and biogeochemical cycles, western Pacific

Sep 18th, 2013 – Oct 2nd, 2013

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

•Contents

- 1. Cruise information
- 2. Researchers
 - 2.1 On board scientists
 - 2.2 Shinkai 6500 operation team
 - 2.3 Crew members
- 3. Observation
 - 3.1.General purpose
 - 3.2. In situ experiments to reveal benthic activities and biogeochemical cycles on the seafloor Hidetaka Nomaki and YK13-09, YK13-12 scientists
 - 3.3. Significance of nitrification in oligotrophic deep sea sediments
 - Takuro Nunoura, Manabu Nishizawa
 - 3.4. Distribution, genetic diversity and food preferences of Xenophyophorean foraminifers in the abyssal plain
 - Masashi Tsuchiya, Hidetaka Nomaki
 - 3.5. Unveiling the drivers of virus-prokaryotes activity and interactions in abyssal ecosystems
 - Eugenio Rastelli and Bruna Petani
 - 3.6. Acknowledgment
 - 3.7. List of observation equipments
 - 3.8. Cruise log
- 4. Notice on using

1. Cruise Information

- Cruise ID: YK13-09
- Name of vessel: R/V Yokosuka
- Title of the cruise: In situ experimental & sampling study to understand abyssal biodiversity and

biogeochemical cycles

- Title of proposal: *In situ* experimental & sampling study to understand abyssal biodiversity and biogeochemical cycles
- Cruise period: 18th Sep to 2nd Oct, 2013
- Ports of call: Guam to Fiji
- Research area: western equatorial Pacific
- Research Map



2. Researchers

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.]

Takuro Nunoura [JAMSTEC] Masashi Tsuchiya [JAMSTEC] Yuki Uejima [Kumamoto University] Eugenio Rastelli [Polytechnic University of Marche] Bruna Petani [Polytechnic University of Marche] Toshimasa Nasu [Nippon Marine Enterprise]

2.2. Shinkai 6500 operation team

Submersible Op. Manager	Kazuhiro Chiba
1st Submersible Staff	Kazuki Iijima
1st Submersible Staff	Mitsuhiro Ueki
1st Submersible Staff	Keita Matsumoto
2nd Submersible Staff	Hirofumi Ueki
2nd Submersible Staff	Fumitaka Saito
2nd Submersible Staff	Takuma Onishi
3rd Submersible Staff	Yudai Tayama

2.3 Crew members

Captain	Shinya Ryono
Chief Officer	Naoto Kimura
2nd Officer	Tetsuo Shirayama
3rd Officer	Hiroharu Omae
Chief Engineer	Eiji Sakaguchi
1st Engineer	Kazunori Noguchi
2nd Engineer	Kenichi Shirakata
3rd Engineer	Kota Kataoka
Chief Radio officer	Masamoto Takahashi
2nd Electronic Operator	Hiroki Ishiwata
3rd Electronic Operator	Ryosuke Komatsu
Boat Swain	Yoshiaki Kawamura

Quarter Master Kazumi Ogasawa		
Quarter Master	Jiro Hanazawa	
Quarter Master	Daizuke Yanagitani	
Sailor	Kazuho Ikeda	
Sailor	Yoshihiro Ogawa	
Sailor	Kenta Nasu	
No.1 Oiler	Kazuaki Nakai	
Oiler	Shinya Sugi	
Oiler	Masayuki Fujiwara	
Oiler	Tatsuomi Chino	
Oiler	Toshinori Matsui	
Chief Steward	Sueto Sasaki	
Steward	Shinsuke Tanaka	
Steward	Masanao Kunita	
Steward	Kazuma Sonoda	
Steward	Shiho Shimizu	

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. 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 different surface productivities.

3.2. In situ experiments to reveal benthic activities and biogeochemical cycles on the seafloor

Hidetaka Nomaki and YK13-09, YK13-12 scientists

We carried out 3 different in situ incubation experiments together with sediment geochemistry investigation at the 1N site (1°15N, 163°15E, water depth = 4277m) during dives #1367 and #1368 on 24th Sep and 26th Sep, respectively. The first incubation is to reveal the chemoautotrophic carbon production rate at the deep-sea floor using in situ incubation cores. The 2^{nd} incubation is to reveal consumption of diatom and cyanobacteria by heterotrophic organisms in sediments using in situ incubation boxes. The third incubation is to reveal the effects of holothurians on microbial community and consumption rates of diatom and cyanobacteria using holothurians cages. All of the incubation were carried out for 2 days incubation and ~50 days incubation, which will be retrieved during YK13-12 cruise.

We collected sediment samples with push corer, water samples with Niskin water sampler, and megabenthos samples with slurp gun (suction sampler).

On board, sediment samples were sliced into different depth layers and subsampled for viral, microbial, geochemical, and meiofaunal analyses. Water samples were filtered on board and the filters were kept frozen. Megabenthos samples were preserved with either ethanol, formalin, or kept frozen.

3.3. Significance of nitrification in oligotrophic deep sea sediments

Takuro Nunoura, Manabu Nishizawa (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 north pacific gyre and equatorial pacific sites, 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.

3.4. Distribution, genetic diversity and food preferences of Xenophyophorean foraminifers in the abyssal plain

Masashi Tsuchiya, Hidetaka Nomaki (JAMSTEC)

Background

Although foraminifers form an ecologically important link between bacteria and macrobenthos in biological and physical cycles in nature, not enough studies have been conducted to clarify this. Studies on protists are indispensable to clarify the biological diversity of the deep-sea floor.

A large unicellular foraminifera, Xenophyophore, have large cell up to $10 \sim 15$ cm in diameter, making their body with reticulate, bush or fan-like structures. Several studies have been conducted for classification and time-lapse observation (e.g. Gooday et al. 1993). Recently, molecular phylogenetic studies and ultrastructural observation were carried out for Xenophyophore, *Shinkaiya lindsayi* (Lecroq et al. 2009). Interestingly, they have two kinds of cytoplasm, stercomare and granellae, although they are unicellular eukaryotes that accumulate heavy metals (Swinbanks and Shirayama 1986, Lecroq et al. 2009). However, the accumulation mechanisms of heavy metals and the ecological role of Xenophophore in the deep-sea are not clear.

Purpose

This study aims to clarify the genetic/environmental background of heavy metal accumulation and the ecological roles of Xenophyophore in the deep-sea population by using molecular techniques and stable isotope measurements. In this cruise, we collected Xenophyophore to illustrate their food preferences. We also carried out visual observation from the Shinkai 6500 to understand the distribution and density of Xenophyophores.

Research results

- 1) Dive #1367 and 1368 were conducted at the 1N site (1°15.0'N, 163°14.8'E, 4,277 m).
- 2) Visual observation were done for landscape, sediment facies, distribution of organisms, and carried out sampling.
- Sampling of sediment cores for SI analyses and DNA analyses, stored in -80°C. Sampling of four types of Xenophyophores

3.5. Unveiling the drivers of virus-prokaryotes activity and interactions in abyssal ecosystems

Eugenio Rastelli and Bruna Petani

(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 YK13-09 cruise organised by JAMSTEC (09-2013), 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) and Japan trench (12-2012). 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.

Materials and methods: Seawater samples were collected during the YK13-09 cruise in station 1N, from

the sea surface and the bottom of the abyssal ecosystem in the pacific ocean. Surface and sub-surface sediments were collected from the same station.

Additional experiments (*in situ* temperature incubations of replicated mesocosms) were conducted on board and *in situ* for sediment samples.

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 Prokaryotic heterotrophic production; extracellular enzymatic activities; organic matter composition and turnover; prokaryotic benthic diversity through next generation sequencing; DNA Metagenomics; RNA analysis and transcriptomics.

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

Future work: The comparison of the new results with those obtained previously during JAMSTEC cruises in the Ogasawara trench and Abyssal plain (2011) and Japan trench (2012) and from future cruises (Mariana Trench, 2014) will allow to gain more information about the functioning of the trench ecosystem in relation to the surrounding abyssal environment. Discussion and collaboration with the other groups of scientists will lead to a more complete knowledge of the connectivity between different trenches and will shed new lights on the deep-sea ecology and ecosystem functioning of these areas.

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. 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.

3.7. 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) H-type push corer (inner diameter: 8.2cm, length: 32cm) Suction sampler with multiple canister

3.8. Cruise log

Date	Local Time	Description	Note	Position/Weather/Wind/Sea condition
17-Sep-13		Scientists onboard.		9/17 12:00 (UTC+10h)
	13:30	Scientists onboard.		APRA bay
	15:00-16:30	Carried out Scientists meeting.		13-27.7N,144-40.0E
				Fine but cloudy
				NW-4 (Moderate breeze)
				2 (Sea smooth)
				1 (Low swell sea)
				Visibly: 8'
18-Sep-13		Let go all shore lines & left SUVA for Research area.		9/18 12:00 (UTC+10h)
	10:00	Let go all shore lines & left SUVA for Research area(OFF East Guam).		OFF West Guam
	11:00-11:30	Carried out shipboard education & training for scientists		13-44.8N,144-48.4E
	13:00-13:30	Carried out SHINKAI 6500 team & Scientists meeting.		Cloudy
	16:40-17:00	Carried out KONPIRA pray.		NW-3(Gentle breeze)
				3 (Sea slight)
				3(Moderate short)
				Visibly: 8'
19-Sep-13		Proceeding to research area.		9/19 12:00 (UTC+10h)
	08:30-09:00	Carried out Scientists meeting.		OFF East GUAM
				12-00.0N,149-15.0E
				Overcast
				SW-6 (Strong Breeze)
				4 (Sea moderate)
				4 (Moderate long)
				Visibly: 6'
20-Sep-13		Arrived at research area(OFF east Guam).		9/20 12:00 (UTC+10h)

	09:30	Arrived at research area(OFF east Guam).		OFF East GUAM
	09:40	Carried out released XBT		12-03.0N,154-00.0E
	09:50	Com'ced surface water samplinng.	12-01.1448N,153-48.9850E	Cloudy
	09:55	Finished surface water samplinng.	12-01.2068N,153-49.0577E	South-4 (Moderate breeze)
	10:00	Com'ced towing proton magnetometer.		4 (Sea moderate)
	11:16-12:02	Carried out MBES site survey.		4 (Moderate average)
	13:11	Com'ced MBES mapping survey.		Visibly: 8'
21-Sep-13		Suspended SHINKAI 6500 submergence due to rough		9/21 12:00 (UTC+10h)
21 Sep 13		sea.		9/21 12:00 (010+101)
	04:50	Finished MBES mapping survey.		OFF North-West POMPEI
	05:30	Suspended SHINKAI 6500 submergence due to rough sea.		11-08.5N,154-47.8E
	06:03	Recovered proton magnetometer.		Cloudy
	06:10	left YOKOSUKA for Research area(OFF East Guam).		NW-3 (Gentle breeze)
		& Com'ced shifting to next research area(Vicinity of the		3 (Sea slight)
		Equator).		3 (Sea slight)
				4 (Moderate average)
				Visibly: 8'
22-Sep-13		Transit to research area		9/22 12:00 (UTC+10h)
	13:00-14:00	Scientists meeting.		OFF North POMPEI
	24:00	shift local time to UTC+11h.		07-23.7N,158-15.5E
				Fine but cloudy
				SSE-3 (Gentle breeze)
				2 (Sea smooth)
				2 (Low swell long)
				Visibly: 8'
23-Sep-13		Transit to research area		9/23 12:00 (UTC+11h)
	11:30-11:50	Scientists meeting.		OFF North-West Naul
	13:00-13:30	6K team & Dive Scientists meeting.		03-34.2N,161-23.1E
				Fine but cloudy
				SE-2 (Light breeze)
				2 (Sea smooth)
				1 (Low swell sea)

				Visibly: 8'
24-Sep-13		SHINKAI6500 Operation Dive#1367		9/24 12:00 (UTC+11h)
	04:20	Arrived at research area(Vicinity of the Equator).		OFF West-North-West
	04-20	Arrived at research area vicinity of the Equatory.		Naul
	04:35	Released XBT		01-15.0N,163-15.0E
	05:00-05:47	Carried out MBES site survey.		Fine but cloudy
	09:04	SHINKAI6500 dove & started her operation #1367.		SSW-3 (Gentle breeze)
	10:56	SHINKAI 6500 landed on sea bottom ($\mathrm{D}\text{=}4277\mathrm{m}$).		2 (Sea smooth)
	15:16	SHINKAI 6500 left sea bottom (D=4254m).		2 (Low swell long)
	16:56	SHINKAI 6500 refloated.		Visibly: 8'
	17:23	Recovered SHINKAI 6500 & finished above operation.		
	17:55	Com'ced towing proton magnetometer.		
	20:14	Com'ced MBES mapping survey.		
25-Sep-13		Carried MBES mapping suevey.		9/25 12:00 (UTC+11h)
	08:15-09:00	scientific meeting		OFF North-West Naul
	11:23	Recovered proton magnetometer.		01-14.8N,163-16.1E
	11:35	Com'ced surface water samplinng.	01-15.0075N,163-15.0063E	Fine but cloudy
	11:41	Finished surface water samplinng.	01-15.0044N,163-14.8917E	SE-2 (Light breeze)
	11:44	Com'ced towing proton magnetometer.		2 (Sea smooth)
				2 (Low swell long)
				Visibly: 8'
26-Sep-13		SHINKAI6500 Operation Dive#1367		9/26 12:00 (UTC+11h)
	05:10	Failed MDEC		OFF West-North-West
	05:16	Finished MBES mapping survey		Naul
	06:06	Recovered proton magnetometer.		01-14.8N,163-16.1E
	09:00	SHINKAI6500 dove & started her operation #1368.		Fine but cloudy
	10:55	SHINKAI 6500 landed on sea bottom (D=4277m).		SE-2 (Light breeze)
	12:58	Com'ced surface water samplinng.	01-15.0277N,163-14.7608E	2 (Sea smooth)
	13:02	Finished surface water samplinng.	01-15.0375N,163-14.7334E	2 (Low swell long)
	15:16	SHINKAI 6500 left sea bottom (D=4255m).		Visibly: 8'
	16:52	SHINKAI 6500 refloated.		

	17:17	Recovered SHINKAI 6500 & finished above operation.	
	17:50	Com'ced proceeding to SUVA.	
27-Sep-13		proceeding to SUVA.	9/27 12:00 (UTC+11h)
	13:30-14:00	Scientists meeting.	OFF South-West Naul
			01-58.6S,165-37.1E
			Fine but cloudy
			ENE-3 (Gentle breeze)
			2 (Sea smooth)
			1 (Low swell sea)
			Visibly: 8'
28-Sep-13		proceeding to SUVA.	9/28 12:00 (UTC+11h)
	00:00 10:00	Onboard scientist seminar.	OFF North-East SANTA
	09:00-10:00	Onboard scientist seminar.	CRUZ
			05-58.6S,165-37.1E
			Fine but cloudy
			WNW-2 (Light breeze)
			2 (Sea smooth)
			1 (Low swell sea)
			Visibly: 8'
29-Sep-13		proceeding to SUVA.	9/29 12:00 (UTC+11h)
	13:00	Practiced boat station drill	OFF East SANTA CRUZ
			09-37.1S,171-15.9E
			Cloudy
			East-5 (Fresh breeze)
			3 (Sea slight)
			1 (Low swell sea)
			Visibly: 8'
30-Sep-13		proceeding to SUVA.	9/30 12:00 (UTC+12h)
			OFF North-West FIJI
			12-56.9S,173-12.4E
			Fine but cloudy

			SE-5 (Fresh breeze)
			4 (Sea moderate)
			3 (Moderate short)
			Visibly: 8'
1-Oct-13		proceeding to SUVA.	10/1 12:00 (UTC+12h)
	18:00-18:30	Scientists meeting.	OFF West FIJI
			17-15.4S,175-31.7E
			Fine but cloudy
			SE-6 (Strong breeze)
			4 (Sea moderate)
			3 (Moderate short)
			Visibly: 8'
2-Oct-13		Arrived at SUVA	
	09:00	Sent out 1st shore line, arrived at SUVA,conpleted YK13-09.	

4. 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.