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PRELIMINARY REPORT

FOR YOKOSUKA

Cruise no. YK 10-16

Challenger deep

Mariana trench, Pacific Ocean

November 20 – December 6, 2010

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

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1. Cruise Information

 # Cruise number: YK10-16

 # Ship name: R/V YOKOSUKA

 # Title of the cruise: Biogeosciences at the Challenger Deep, the deepest point of the world: relict organisms and their relations to biogeochemical cycles

 # Chief scientist: Hiroshi Kitazato (Institute of Biogeosciences, Japan Agency for Marine-Earth Science and Technology)

 # Representative of Science Party: Hiroshi Kitazato (Institute of Biogeosciences, Japan Agency for Marine-Earth Science and Technology)

 # Science Party on board: Hiroshi Kitazato, Kazumasa Oguri, Takashi Toyofuku, Hidetaka Nomaki, Beatrice Lecroq (BioGeos, JAMSTEC), Ronnie N. Glud, Anni Glud (Univ. Southern Denmark), Mathias Middelboe (Univ. Copenhagen), Frank Wenzhoefer (MPI, Bremen)

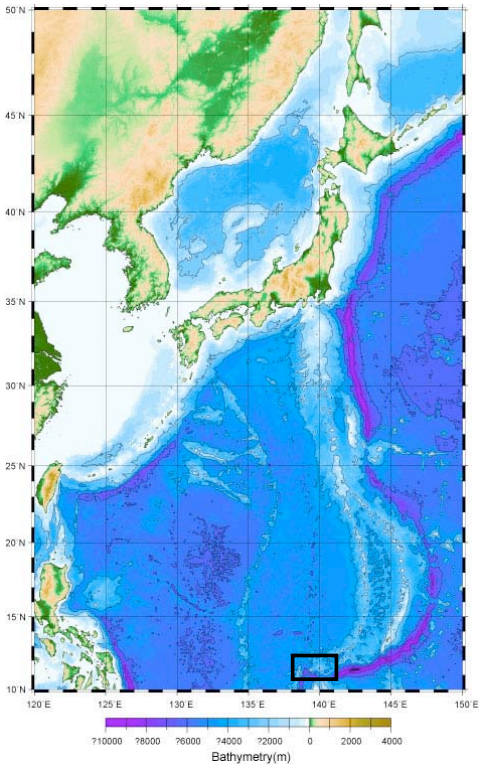
 # Title of proposal: Biogeosciences at the Challenger Deep, the deepest point of the world: relict organisms and their relations to biogeochemical cycles

 # Cruise Period: November 20 ~ December 6, 2010 (from proposal)

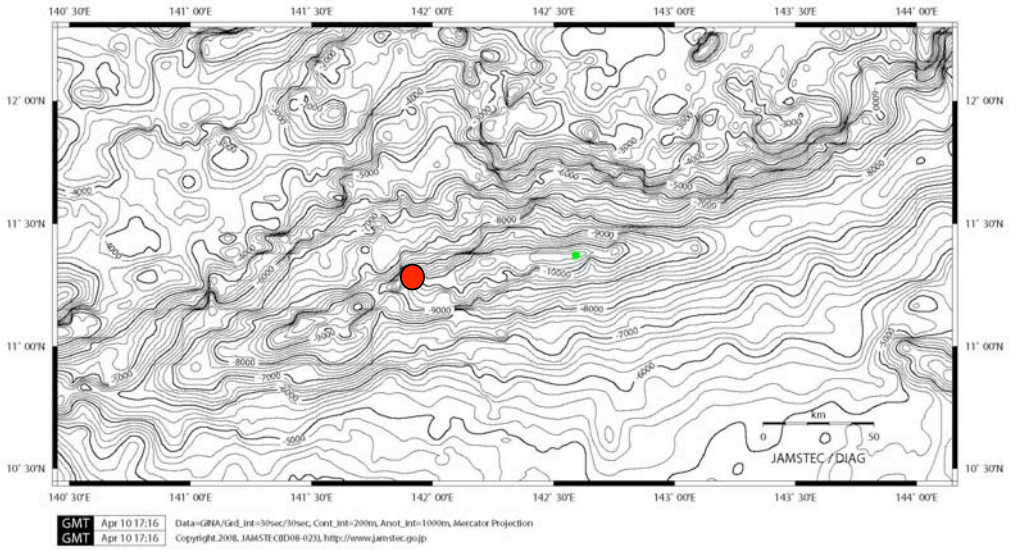
 # Portcall: Apra Harbor, Guam (November 20) ~Apra Harbor, Guam (November 30) ~ JAMSTEC Pier (December 6)

 # Research Area: Challenger Deep area, southern Marianas Trench

 # Research Map:



Bathymetric Map



2. Overview of Research Cruise

Background Idea for the cruise: The Challenger Deep of the Marianas Trench, western Pacific, is the deepest point of the world ocean. We found that simple foraminiferal community flourishes at the deepest point (Todo et al., 2005). More than 99% of foraminiferal community consists of soft-shelled foraminifera. The foraminiferal genera are belonging to phylogenetically ancestral group of the taxon from the morphological point of view. The group is thought to branch in the beginning of the foraminiferal history, 690-1100 Ma according to molecular phylogeny. We would like to know why simple foraminifera flourish at the deepest point of the world. We would like to examine a hypothesis that the deep trenches play acts as refugia for marine organisms.

Purpose: For the purpose to elucidate above hypothesis, we plan to carry out oceanographic researches at the Challenger Deep (c.a. 10,900m deep). The series of researches will be carried out at the deep. They are,

1) Collect both undisturbed sediment cores and macrobenthos samples by free-fall type coring devices / camera / CTD / baited trap system.
2) Measure sediment-water interface O2 profiles with microelectrodes that are assembled on ultra-deep lander system at the Challenger Deep to estimate oxygen consumption rate in the deep trenches.
3) Make DNA / RNA analyses for both foraminifera in cores and macrobenthos in the baited trap samples for molecular phylogeny. Environmental DNA will also be analysed by the massive sequencing method.

For this purpose, we use two mooring systems, free-fall camera / CTD / coring system and ultra deep lander system.

Description of researches: Researches were carried out at two depths, both 6000m and 10850m. Shallower site is located at ocean-side slope of the Challenger Deep. Deeper site locates at western deep of the Challenger Deep where the deepest depth is recorded.

3. Onboard Researchers and Crews:

Chief Scientist on board: Hiroshi Kitazato (BioGeos, JAMSTEC)

Representative of Science Party: Hiroshi Kitazato (BioGeos, JAMSTEC)

Science Party:

No.	Name	Affiliation	Research field
1	Kitazato, Hiroshi	Biogeos, JAMSTEC	Biogeosciences and cruise coordinator
2	Oguri, Kazumasa	Biogeos, JAMSTEC	Sedimentology
3	Glud, Ronnie	University of Southern Denmark	Biogeochemistry/Microbiology
4	Glud, Anni	University of Southern Denmark	Biogeochemistry/Microbiology
5	Middelboe, Mathias	University of Copenhagen	Biogeochemistry/Microbiology
6	Wenzhoefer, Frank	Max Planck Institute	Biogeochemistry/Microbiology
7	Nomaki, Hidetaka	Biogeos, JAMSTEC	Deap-sea ecology
8	Toyofuku, Takashi	Biogeos, JAMSTEC	Geobiology
9	Lecroq, Beatrice	JSPS/Biogeos, JAMSTEC	Marine Biology
10	Ogura, Satoshi	Nippon Marine Enterprises	Operation and management of Technical support
11	Miura, Atsumori	Nippon Marine Enterprises	Technical support
12	Onishi, Takuma	Nippon Marine Enterprises	Technical support
13	Asai, Ryu	Nippon Marine Enterprises	Technical support

Affiliations:

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

Yokosuka Headquaters

University of Southern Denmark

Biological Institute

University of Copenhagen

Department of Biology

Max-Planck Institute for Marine Microbiology

Celsiusstraße 1

Ship Crew

1	Susami, Satoshi	Captain
2	Kimura, Naoto	Chief Officer
3	Ohara, Toshiyo	2nd Officer
4	Kobayashi, Yumihiko	3rd Officer
5	Shiojima, Tsubasa	Jr. 3rd Officer
6	Kimura, Toshihiro	Chief Engineer
7	Noguchi, Kazunori	1st Engineer
8	Shirakata, Ken'ichi	2nd Engineer
9	Ikeguchi, Kenta	3rd Engineer
10	Nagano Shota	Jr. 3rd Engineer
11	Takahashi, Masamoto	Chief Electronics Officer
12	Ishiwata, Hiroki	2nd Electoronics Officer
13	Minamoto, Mai	3rd Electoronics Officer
14	Abe, Kazuo	Boat swain
15	Kadoguchi, Kuniharu	Able Seaman
16	Oda, Hatsuo	Able Seaman
17	Yoshino, Yuki	Able Seaman
18	Iwasaki, Naoki	Able Seaman
19	Tamura, Ryoma	Sailor
20	Miyashita, Takuya	Sailor
21	Miura, Shun	Sailor
22	Yahata, Kiyoshi	No. 1 Oiler

23	Kawai, Yoshinori	Oiler
24	Higashigawa, Yuji	Oiler
25	Nakahara, Yuki	Oiler
26	Torao, Shin	Oiler
27	Miyauchi, Takeshi	Chief Steward
28	Chikuba, Yukihide	Steward
29	Sonoda, Kazuma	Steward
30	Fukuda, Hiroki	Steward
31	Nakano Mizuki	Steward

4. Research Cruise

1) Deep-sea amera system

Kazumasa Oguri, Takashi Murashima, Hiroshi Kitazato

To record sediment surface and organisms and to collect sediments, deep-sea camera system was developed (figure 3-1; Murashima et al., 2009). This instrument consists of nineteen deep-sea floats and main aluminum frame mounted transponder/releaser, HDTV camera, light, battery, CTD, three short core samplers and ballast weight (figure 3-2). In this cruise, the camera system was used to collect sediments, sea floor observations, collection of organisms with baited trap, and depth, temperature and salinity measurements in water column. When the camera system was released from the ship, the CTD was started to record pressure, conductivity and temperature. The camera was started to record by the power supply from internal timer circuit. Sediments were collected in the core tubes mounted on the tripod legs during the landing (figure 3-3). The transponder released the ballast weight when receiving the release command from the ship. The position of the camera system was always monitored on board to receiving the acoustic signal from the transponder. Table 3-1 shows the setting of the camera system in each deployment. Figure 3-4 shows the landing sites.

Reference

Murashima, T. Nakajoh, H. Takami, H. Yamauchi, N. Miura, A. Ishizuka, T. (2009) 11,000m class free fall mooring system. Proceedings of OCEANS 2009-EUROPE, 2009. OCEANS ‘09, p1-5.

Table 3-1: Setting of the camera system in each deployment.

	Date deployed	Max. CTD depth (m)	Sediment sampling	HDTV video recording	CTD	Baited Trap
Camera1	2010/11/21	6037				
Camera2	2010/11/22	6038				×
Camera3	2010/11/23-24	No data			Failure	
Camera4	2010/11/25-26	10900				×
Camera5	2010/11/27-28	10900		Failure		×

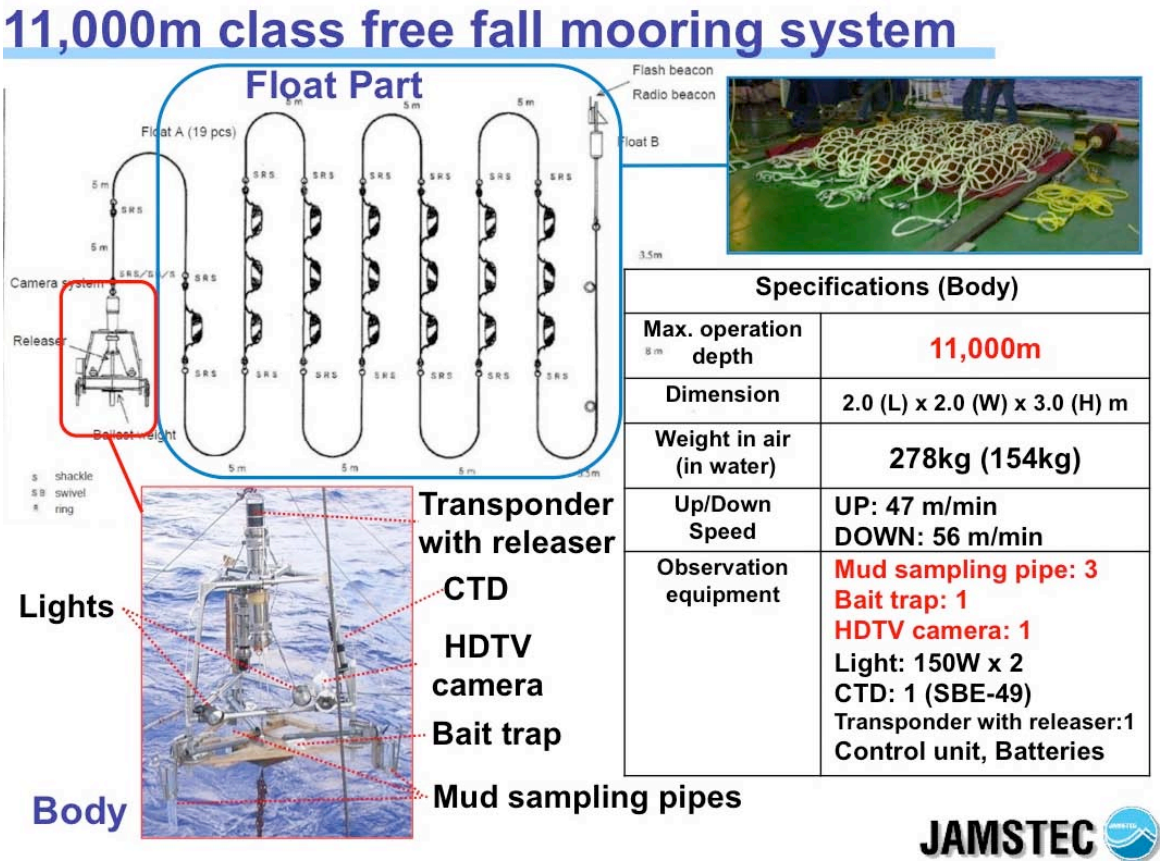


Figure 3-1: Schematic overview of deep-sea camera system.

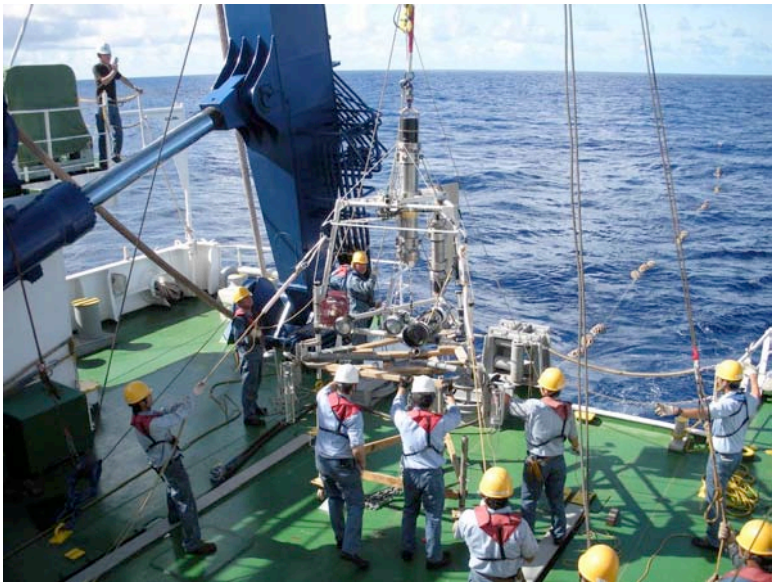


Figure 3-2: Deep-sea camera system (Camera 1).

Figure 3-3: Recovery of the deep-sea camera system (Camera 2). Three sediment cores were collected in each deployment.

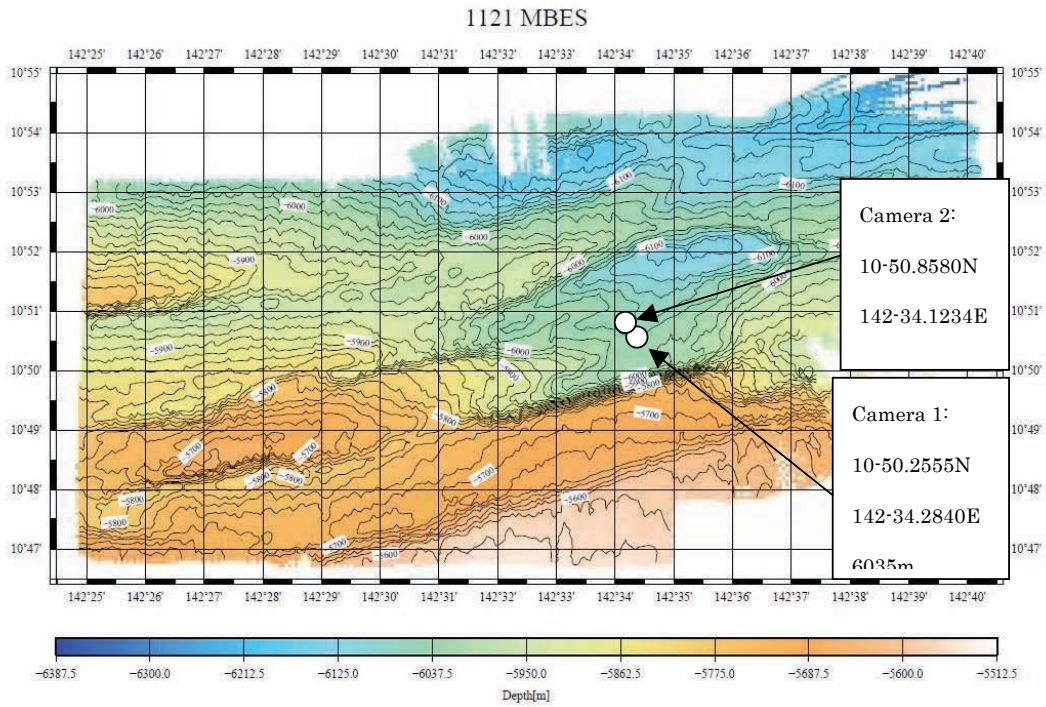


Figure 3-4: Landed points of the deep-sea camera system.

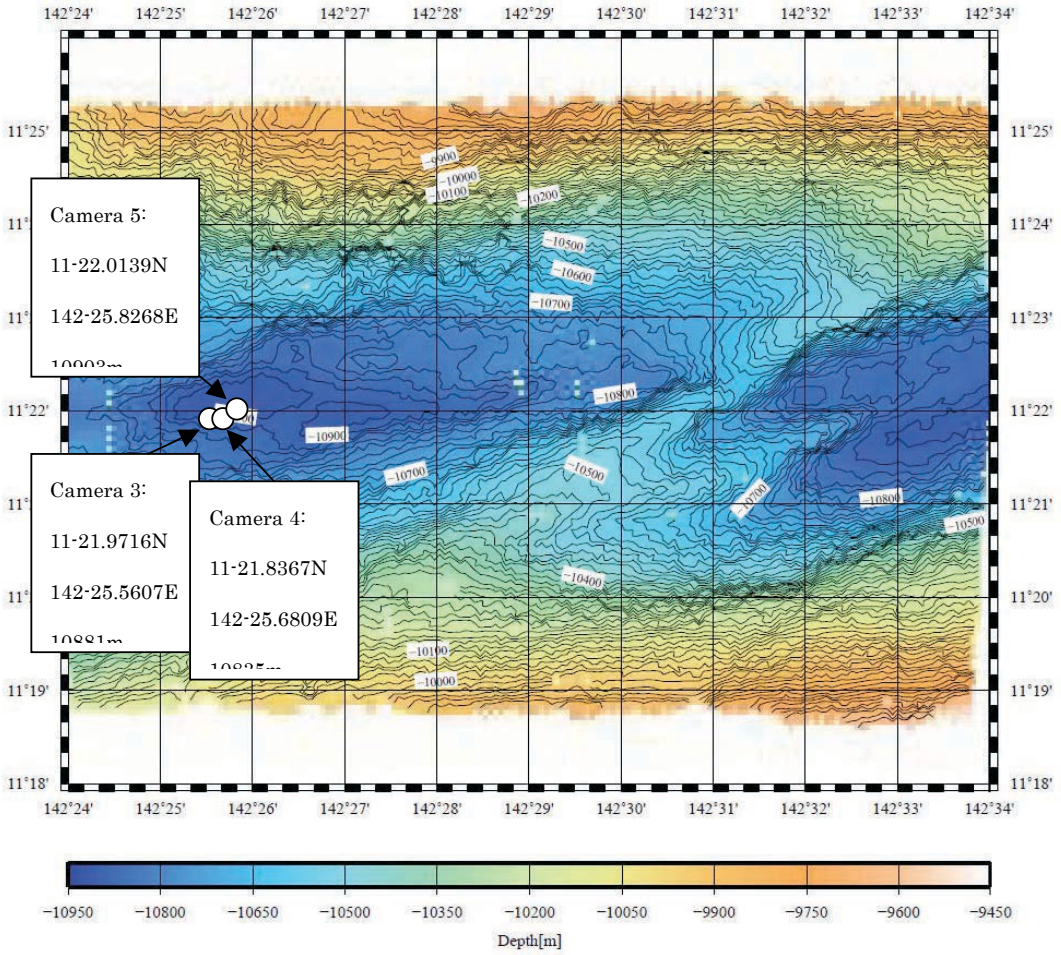


Figure 3-4 continued: Landed points of the deep-sea camera system.

2) Ultra Deep Lander System

Ronnnie N Glud, Frank Wenzhoefer, Mathias Middelboe, Anni Glud

The instrument consist of 4 main components; ballast system, flotation unit, battery and most importantly the measuring unit – additionally two Niskin bottom water samplers were added to the frame.

The instrument is autonomous and once released from the ship it sinks to the sea-bottom by a speed of approx. 45 m min^{-1}. Once settled at the seabed the central measuring unit carry out a pre-programmed measuring routine, before ballast is released and the now positively buoyant system ascend by a speed of approx. 33 m min^{-1}.

The central measuring unit holds an array of microsensors (Fig. 3-5). On this cruise 9 O_2 microelectrodes were mounted at each of the 4 deployments. The thin sensors are lowered into the sediment with a pre-programmed step size (on this cruise 0.5 or 1.0 mm) using a high resolution elevator system. At each step they record a signal proportional to the O_2 content of the sediment. Thereby high resolution vertical microprofiles across the sediment water interface can be resolved. Total profiling distance was set to 25 and 28 cm and after reaching the maximum depth, the sensor array is moved back up to the start position. The entire measuring unit is then moved horizontally and a new vertical measuring routine is initiated. This procedure is repeated as many times as possible at the given deployment time (at this cruise 6-9 times). The ultra deep lander is modified from a previous version that successfully has been deployed in Sagami Bay many times - from *RV Natushima* (Glud et al 2009a, 20009b)

The two added Niskin bottles allow bottom water to be sampled and later onboard the O_2 concentration of this water is determined by Winkler titrations. These values are used for calibrating the in situ measurements along with onboard determination of the “Zero”-current of the sensors.

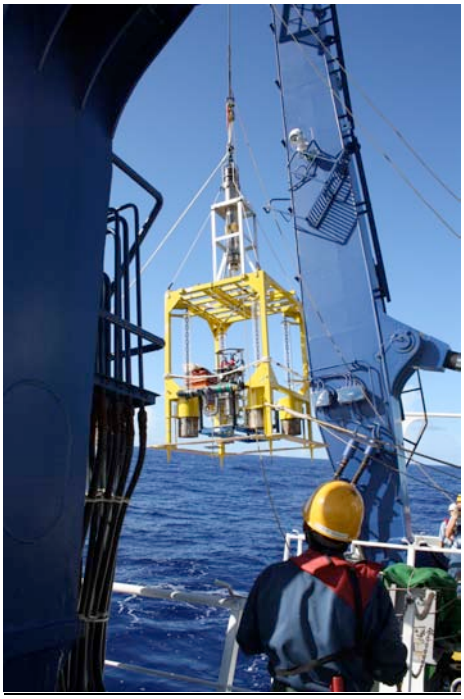


Fig. 3-5: Ultra-deep lander being deployed; ¥horizontal sledge carrying the central electronic unit where the oxygen electrodes are mounted

3) Daily Report

Hiroshi Kitazato

November 20, 2010

Leave Apra Harbor, Guam Island on 11:00 a.m. Steam toward station 6000. We made Kompira-san Praying Ceremony from 16:40-17:00. Around mid-night, we arrived at the 6000m site area and made sea-beam map up to morning.

November 21, 2010

08:00 am start to prepare free-fall camera system. Due to problem of camera shatter system, we should wait c.a. 1 hour for adjustment.

09:40 am deploy camera system into the 6000m deep. (Camera System 1)

11:28 am on bottom at 6032m deep.

14:03 pm send release command.

14:06 pm off bottom

16:10 pm sea surface.

16:50 pm whole system on deck (3 cores, 21 amphipods, CTD?, deep-sea camera photographs)

During the night, sea beam mappings were carried out at the Challenger Deep area.

November 22, 2010

08:00 am stand by for deployment camera system.

08:29 am camera system start down to the bottom (Camera System 2)

10:16 am touched on bottom at 6027m. Ship position is 10-50.41N, 142-33.82E

11:16 am off bottom

13:05 pm surface

13:45 pm on deck. (3 cores)

15:40 pm profiling lander system deployment start. (Lander 1)

17:52 pm lander on bottom (5982m)

Lander position is 10-50.7723’N, 142-33.7676’E, depth 5982m

November 23, 2010

05:46 am send release command

05:53 am off bottom. Upward speed is c.a. 40m / min.

08:40 am surface

09:30 am system on deck. Two niskin bottles were safely closed. Guide pipe is strongly pressed and is difficult to slide the microelectrode system.

13:36 pm free-fall type camera system started down (Camera 3)

16:49 pm on bottom (10,921m)

Calibrated camera system position is 11-21.9358'N, 142-25.6809'E, depth 10,850m.

November 24, 2010

04:59 am off bottom

08:50 am surface

09:39 am on deck. Three cores and two baited traps. Core surface is well oxygenated. In total 64 specimens of scavenging amphipod were collected. Four buoys were broken and changed by new buoys.

13:23 pm profiling lander system deployment started. The system sank down to the Challenger Deep. (Lander 2)

17:17 pm on bottom (10,817m)

Calibrated position is 11-21.9407'N, 142-25.7610'E, depth 10,817m.

November 25, 2010

04:48 am Send release signal

04:53 am Off bottom

08:58 am Surface

09:45 am System on deck. Surface painting films were pieled off under high hydraulic pressures. One bouy has broken. 57 specimens of scavenging amphipods were caught in the baited trap.

14:22 pm Camera system goes down to the Challenger Deep. (Camera 4)

17:35 pm on bottom

Calibrated position is 11-22.1066'N, 142-25.7059'E, depth 10845.7m

November 26, 2010

04:48 am Send release command

04:55 am off bottom

08:25 am surface

09:00 am Camera system on deck. Three cores, CTD, video images.

Cores are processed for massive sequencing, foram faunal study and pick living forams. Soft-shelled foraminifera, such as Resigella, Nodellum and Leptohalysis, are collected for

both DNA analysis and TEM observation.

13:19 pm Lander system goes into the Challenger Deep. (Lander 3)

17:12 pm on bottom

Calibrated position is 11-22.0794'N, 142-25.8253'E, depth 10813m

November 27, 2010

04:59 am send release command

05:06 am off bottom

09:50 am Lander on deck. 42 oxygen profiles and one Niskin bottle. Insitu Winkler titration system was basically worked. But, seawater leaked. In baited trap, 20 scavenging amphipods were trapped.

14:31 pm Camera system goes down into the Challenger Deep. (Camera 5)

17:15 pm on bottom

Calibrated position is 11-21.9810'N, 142-25.8680'E, depth 10889.6m

November 28, 2010

04:46 am send release command

04:59 am off bottom

09:10 am Camera 5 on deck. Three cores and CTD. Video images were not recorded.

13:28 pm Lander system goes into 6000m site. (Lander 4)

15:37 pm on bottom

Calibrated position is 10-50.7612'N, 142-34.0650'E, depth 6018.3m

November 29, 2010

06:13 am send release command

06:15 am off bottom

09:21 am system on deck (oxygen profiles, 2 Niskin bottles, in situ Winkler titration, baited trap (47 scavenging amphipods)

Due to strong wind introduce high waves, camera system deployments for feeding experiments are cancelled. R/V YOKOSUKA steams toward Guam Island.

November 30, 2010

09:00 am portcall at Apra Harbor, Guam. One scientist (Kitazato) and four

operation team members leave R/V YOKOSUKA.

13:00 pm leave from Apra Harbor, Guam and steam toward Yokosuka.

4) Sample List

Observation Record

No.	Recovery Date	Instrument	Site	Latitude			Longitude			depth(m)	Observation variables/Items
1	21-Nov	DCS	CS1	10°	50.3300'	N	142°	33.6900'	E	6032.0	CTD, HD movie , Sediment Corer,Baited trap
2	22-Nov	DCS	CS2	10°	50.8311'	N	142°	34.0585'	E	6070.6	CTD, HD movie , Sediment Corer
3	23-Nov	UDLS	Lander-1	10°	50.7723'	N	142°	33.7676'	E	5982.0	CTD, HD movie , Sediment Corer
4	24-Nov	DCS	CS3	11°	21.9358'	N	142°	25.6809'	E	10850.4	CTD, HD movie , Sediment Corer,Baited trap
5	25-Nov	UDLS	Lander-2	11°	21.9407'	N	142°	25.7610'	E	10817.0	DO profile, Niskin, Baited trap
6	26-Nov	DCS	CS4	11°	22.1066'	N	142°	25.7059'	E	10899.0	CTD, HD movie , Sediment Corer
7	27-Nov	UDLS	Lander-3	11°	22.0794'	N	142°	25.8253'	E	10813.0	DO profile, Niskin, Baited trap, in-situ O2 fixation
8	28-Nov	DCS	CS5	11°	21.9810'	N	142°	25.8680'	E	10900.0	CTD, HD movie , Sediment Corer
9	29-Nov	UDLS	Lander-4	10°	50.7612'	N	142°	34.0650'	E	6018.0	DO profile, Niskin, Baited trap, in-situ O2 fixation

DCS=Deepsea Camera System, UDLS=Ultra Deep Lander System

Lander Observation & Water Sampling

No.	On board ID	Sample/Measurement		locality	Purpose	Ronnie Glud	Anni Glud	Mathias Middelboe	Frank Wenzhoefer	Kazumasa Oguri
1	Lander 1	measurements	electrodes	Mariana Trench	in situ O2 profiles	x	x	x	x	
2	Lander 2	measurements	electrodes	Challenger deep	in situ O2 profiles	x	x	x	x	
3	Lander - W	Sea water	Niskin	Challenger deep	Overlying wqter for DO	x	x	x	x	x
4	Lander 3	measurements	electrodes	Challenger deep	in situ O2 profiles	x	x	x	x	
5	Lander - W	Sea water	Niskin	Challenger deep	Overlying wqter for DO	x	x	x	x	x
6	Lander 4	measurements	electrodes	Mariana Trench	in situ O2 profiles	x	x	x	x	
7	Lander - W	Sea water	Niskin	Mariana Trench	Overlying wqter for DO	x	x	x	x	x

Sediment Samples

No.	On board ID	Sample	locality	Purpose	Hiroshi Kitazato	Hidetaka Nomaki	Beatrice Lecroq	Ronnie Glud	Anni Glud	Mathias Middelboe	Frank Wenzhoefer
1	CS1-1	Sediment	Mariana Trench	for O2, Pb, virus, Chl a, Org C, nematodes				x	x	x	x
2	CS1-2	Sediment	Mariana Trench	for O2, Pb, virus, Chl a, Org C, nematodes				x	x	x	x
3	CS1-3	Sediment	Mariana Trench	for O2, Pb, virus, Chl a, Org C, nematodes				x	x	x	x
4	CS1-W	Sea water	Mariana Trench	Overlying wqter for DO				x	x	x	x
5	CS2 - 1 - 1	Sediment	Mariana Trench	for Foraminifera Assemblage	x						
6	CS2 - 1 - 2	Sediment	Mariana Trench	for foraminiferal massive DNA	x		x				
7	CS2 - 2 - 1	Sediment	Mariana Trench	for Foraminifera Assemblage	x						
8	CS2 - 2 -2	Sediment	Mariana Trench	for foraminiferal massive DNA	x		x				
9	CS2 - 3 -1	Sediment	Mariana Trench	for Foraminifera Assemblage	x						
10	CS2 - 3 -2	Sediment	Mariana Trench	for foraminiferal massive DNA	x		x				
11	CS3 - 1	Sediment	Challenger deep	for O2, Pb, virus, Chl a, Org C, nematodes				x	x	x	x
12	CS3 - 2	Sediment	Challenger deep	for O2, Pb, virus, Chl a, Org C, nematodes				x	x	x	x
13	CS3 - 3	Sediment	Challenger deep	for O2, Pb, virus, Chl a, Org C, nematodes				x	x	x	x
14	CS3 - 4	Sea water	Challenger deep	Overlying wqter for DO				x	x	x	x
15	CS4 - 1- 1	Sediment	Challenger deep	for Foraminifera Assemblage	x						
16	CS4 - 1 - 2	Sediment	Challenger deep	for foraminiferal massive DNA	x		x				
17	CS4 - 2 - 1	Sediment	Challenger deep	for Foraminifera Assemblage	x						
18	CS4 - 2 - 2	Sediment	Challenger deep	for foraminiferal massive DNA	x		x				
19	CS4 - 3 - 1	Sediment	Challenger deep	for Foraminifera Assemblage	x						
20	CS4 - 3 - 2	Sediment	Challenger deep	for foraminiferal massive DNA	x		x				
21	CS5 - 1	Sediment	Challenger deep	for O2, Pb, virus, Org C				x	x	x	x
22	CS5 - 2	Sediment	Challenger deep	for O2, Pb, virus, Org C		x		x	x	x	x
23	CS5 - 3 - 1	Sediment	Challenger deep	for foraminiferal massive DNA			x				

Biological Samples

No.	On board ID	Site	Sample	n=	Method	Storage	locality	Purpose	Investigator
1	CS-1-trap1	CS1	Amphipod	21	Baited trap	Frozen	Mariana Trench	Biometry & DNA extraction	Watanabe
2	CS-1-trap2	CS1	Amphipod	1	Baited trap	Frozen	Mariana Trench	Biometry & DNA extraction	Watanabe
3	CS-2-sed3	CS2	Soft shelled foraminifera	ca. 10	Deep-sea Camera Corer	Frozen	Mariana Trench	DNA extraction	Kitazato
4	CS-3-lower	CS3	*Hirondellea gigas*	60	Baited trap	Frozen	Challenger deep	Biometry & DNA extraction	Watanabe
5	CS-3-upper	CS3	*Hirondellea gigas*	4	Baited trap	Frozen	Challenger deep	Biometry & DNA extraction	Watanabe
6	Lander 2-trap	Lander 2	*Hirondellea gigas*	57	Baited trap	Frozen	Challenger deep	Biometry & DNA extraction	Watanabe
7	CS-4-sed3-1	CS4	Soft shelled foraminifera	ca. 15	Deep-sea Camera Corer	Frozen	Challenger deep	DNA extraction	Kitazato
8	CS-4-sed3-2	CS4	Soft shelled foraminifera	ca. 100	Deep-sea Camera Corer	Glutaraldehyde fixation	Challenger deep	TEM observation	Kitazato
9	Lander 3-trap	Lander 3	*Hirondellea gigas*	20	Baited trap	Frozen	Challenger deep	Biometry & DNA extraction	Watanabe
10	Lander 4-trap	Lander 4	Amphipod (rounded)	11	Baited trap	Frozen	Mariana Trench	Biometry & DNA extraction	Watanabe
10	Lander 4-trap	Lander 4	Amphipod (elongated)	37	Baited trap	Frozen	Mariana Trench	Biometry & DNA extraction	Watanabe

5. Research Proposals

1) Foraminifera in the Challenger Deep

Hiroshi Kitazato, Beatrice Lecroq,
Nina Ohkawara and Andrew J. Gooday
(BioGeos, JAMSTEC and NOCS, UK)

Introduction

What kinds of organisms do dwell in the deepest point of the world ocean? This is a simple question. In 2005, we published a quick report in terms of species composition of foraminifera that dwell in the Challenger Deep (Todo et al., 2005). The fauna was composed of only 13 species. Soft-shelled foraminifera occupy 99% of the fauna. The lineage of soft-shelled foraminifera are thought to be traced back to the pre-Cambrian Era (Pawlowski et al., 2003). For getting a proof in terms of this quick report, we would like to get duplicate samples for molecular, statistical and cytological analyses.

Method of study

Surface undisturbed core samples are planned to use for foraminiferal research. Not only we shall make researches on faunal analyses according to external morphological characters, but also we plan to analyse species level DNA phylogeny and observe cell anatomy by TEM. Syringe subcores are inserted into each core. Each subcore will be sliced every 0.5 cm intervals from core top to 3 cm and every 1 cm intervals for 3 to 5 cm. From the slices, we pick individuals for the researches. After washing through wet sieves with 32 μm opening, we sort out living individuals both for DNA analyses and for TEM observations. For the faunal analyses, every sliced fractions are fixed with 1% sea water- Rose Bengal formalin solution. Just before sorting, we plan to wash fractions with 32 μm opening mesh. Then, every stained individuals will be sorted at species level under binocular stereo-microscope.

Procedures on board

During the CS-2 and -4 deployment of YK10-16 cruise, we collected three cores with free-fall type camera system. We took three subcore samples from each of cores with 50 ml plastic syringe. One subcore samples are sliced with core slicer and fixed with rose Bengal formalin solution. Another subcore is used for picking live

foraminifera. We picked 100 specimens belonging to six morphospecies, *Resigella biloculata, R. laevis, Nodellum aculeate, N. cf. aculeate, Lagenammina* sp. and *Reophax* sp. These species have already reported from the Challenger Deep area (Gooday et al., 2008a,b ; Kitazato et al., 2009). All specimen were fixed with glutaraldehyde solution, and then were rinsed four times with filtered bottom water. The fixed specimens were stored in a refrigerator until after-fix procedure will be made. Specimens for species level DNA study were frozen with liquid nitrogen and stored in -80 °Cdeep-freezer.

Future study

Faunal analyses will be carried out in species level. Cytological observation for fixed specimens will be carried out with TEM. Species level DNA analyses will be made. All the procedures and analyses plan to be done at JAMSTEC laboratory.

Potential collaborators

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2) Ultra Deep Lander System

Ronnnie N Glud, Frank Wenzhoefer, Mathias Middelboe, Anni Glud_

Background

The amount of organic material that escapes mineralization and is retained in the sediment record is the single most important factor determining the O_2 levels of the global ocean and our planet. Today we have a reasonable good understanding of the processes that are responsible for mineralization of organic material and which factors regulate the efficiency by which organic material is degraded in the seabed. However, to obtain firm quantitative measures we need to conduct direct measurements in the respective areas of our ocean. The current global data base of high-quality measurements of benthic carbon mineralization rate is around 400 spread over the global ocean (Glud 2008). This limited database is mainly concentrated to a few regions of the global ocean and there exist large "white", unexplored areas of the seabed – including the deep trenches. Trenches only account for less than 2% of the global seabed area, but could via sediment focusing act as traps for organic material. Consequently the mineralization efficiency at these great depth could play a crucial role for the global carbon (and O_2) household. *We hypothesize that trenches play an un-proportional large role for retention of organic material in the sea-bed.*

The benthic mineralization is mainly driven by vast numbers of Bacteria and Archaea. Currently even the most basic information on abundance and distribution of microbes in trench sediments are missing. One of the main mortality factors for microbes in deep-sea sediments are deadly virus infections (Danovaro et al 2008) and bacterial cell walls appear to be a major component of relict marine organic material (Lomstein et al 2009) . Currently there exists no knowledge about the role of viruses for regulating the mortality or the diversity of microbes in trench sediments, or in deep sea sediments in general.

We want to quantify the carbon mineralization efficiency at Challenger deep (the deepest site on Earth) and compare it to conditions at the neighbouring abyssal plain –

further we wish to quantify the distribution and diversity of prokaryotes and virus in these sediments.

Strategy

We measured a number of O_2 microprofiles at Challenger deep and at the abyssal plain using the Ultra-deep lander system. From these profiles we can calculate how much O_2 is consumed in the sediment and also where it is consumed within the oxic zone. This provides indications on which processes are responsible for the benthic O_2 consumption. The O_2 consumption rate provides an integrated measure of the total benthic carbon mineralization rate. Profiles measured in situ will be compared to the more commonly applied laboratory based measuring procedures to resolve the extent by which such profiles are affected by recovery artefacts (Glud et al 1999, Hall et al 2007.

Recovered sediment cores were sectioned and the respective slices will be used for dating by the ^{210}Pb procedure. Combined with information on the distribution of organic material (and the C/N ratio) this will allow us to quantify the sediment accumulation rates and the retention of carbon and nitrogen in the sediment record. Thereby the mineralization efficiency can be determined at the investigated sites. Important back-ground information on sediment density and porosity that are required for the calculations will also be determined. Further, complementary information on sediment accumulation (and mixing) will be obtained from depth profiles of phototrophic pigments stored in the sediment. A few samples for investigating nematods and analids were taken for colleagues back home.

The vertical distribution of Bacteria (+ Archea) and virus will be determined by an optimized extraction, staining and counting procedure (Middelboe and Glud 2006, Siem-Joergensen et al 2008). Further, virus and bacteria biomass will be concentrated to perform metagenomic analyses (Breitbart et al 2002) in 6 sediment depths.

To sites were investigated and will be compared; the Challenger Deep at a depth of ~10.800m and a site outside the trench at ~6.000 m water depth.

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3) Foraminiferal richness assessment using second generation massive sequencing

Lecroq Beatrice, Hiroshi Kitazato

Purposes

This cruise is part of a project aiming to increase our knowledge of morphological, molecular and ecological diversity of deep-sea benthic foraminifera.

For massive sequencing analyses strategy, the specific objective of this field campaign was to collect sediment samples from Challenger Deep, Marianna Trench in Pacific Ocean to answer the three following questions:

- What is the foraminiferal richness of the sediment at the deepest point of earth and how accurate can it be retrieved from environmental massive sequencing method?
- What is the metabolic activity of the foraminiferal communities (reflected by environmental RNA)?
- What is the spatial variation of foraminiferal richness at micro- and meso-scale on the surface of the sediment and down to the deeper 40 cm layers?

Background

Deep-sea sediments are home for a wide range of small-sized metazoan and protistan taxa. The diversity of this meiofaunal community is difficult to estimate, since its study suffers from undersampling, difficult access and impossibility to cultivate deep-sea organism. Moreover, most of the deep-sea species are tiny, fragile and difficult to identify. Benthic foraminifera form the most abundant and diverse group of deep-sea meiofauna, found even in the deepest ocean trenches. It has been shown that the large proportion of the deep-sea foraminifera belong to the early lineages characterized by simple, single-chamber (monothalamous), organic-walled or agglutinated tests, poorly preserved in the fossil record. Because of the poor preservation of their tests, their small size and the lack of distinctive morphological features, their diversity is especially

difficult to assess. The environmental massive sequencing approach consisted in getting numerous sequences from the global DNA extractions from environmental samples overcoming the time-consuming step of species isolation and identification under microscope and providing by far more sequence data than the traditional cloning method.

Methods and samples processing on board

The sediment samples for foraminiferal studies were collected using the Shinkai Camera System coupled with 3 push cores of 74 mm in diameter and 60 cm long. Samples have been collected at stations CS2, CS4 and CS5 (for further information on sampling sites please refer to the stations list). For stations CS2 and CS4, replicates have been performed using sediment from each of the 3 cores from the same dive in order to make statistical analyses and investigate the micro- and meso-scale variation in the richness composition. For both stations CS2 and CS4, subsampling from the same cores has been performed for 1) DNA/RNA massive sequencing analysis, 2) faunal studies and 3) observation of isolated living forminifera specimen.

After deployment and recovering on deck, cores were immediately placed in cold place to prevent deterioration of DNA and RNA material. Subsampling for massive sequencing purpose has been conducted in sterile conditions and consisted in aliquoting 1 or 2 mL of raw sediment from each cm layer up to 20 cm, 40 cm and 5 cm for CS2, CS4 and CS5 respectively. Each aliquot was immediately deep frozen into liquid nitrogen before getting stored in deep freezer at -80°C.

4) **Genetic diversity, connectivity and trophic characteristics of a giant amphipod *Hirondellea gigas* in the Mariana Trench**

Hiromi WATANABE, Katsunori FUJIKURA, Hidetaka NOMAKI

The ecology of deep-sea animals is not understood so much, mainly due to inaccessibility to deep sea. Genetic information and other chemical features of the animals, such as stable isotope composition, sometimes provide an overview of ecological characteristics of the animals. A giant amphipod *Hirondella gigas* has been collected in the deepest marine environment such as trench. In the present study, we use genetic and stable isotopic analyses to estimate their effective population size difference, gene flow and trophic relationships among vertically distant populations in the Mariana Trench.

To collect the giant amphipods in the harsh environment, bate trap (Figure) was deployed with the lander system. Totally 61 individuals of the giant amphipod were collected from the Mariana Trench, a single individual at the 6000m site and 60 individuals at the Challenger Deep. In addition, about 20 individuals of another species of amphipod were also collected at the 6000m site. These amphipods were frozen on board.

The frozen amphipods will be used for DNA extraction and stable isotopic analyses. Both of mitochondrial and nuclear DNA sequences of these amphipods will be obtained and analyzed for the population-level diversity and connectivity. From these analyses, we will infer reproductive and dispersal strategies of these amphipods in the Mariana Trench.

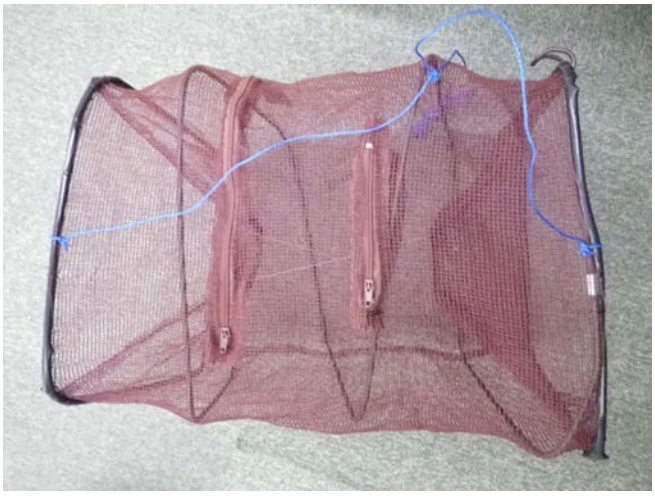


Figure 5-1: Bate trap in YK10-16 cruise. A chopped scombrid mackerel was used as a bate in each trap.

6. Data

1) CTD

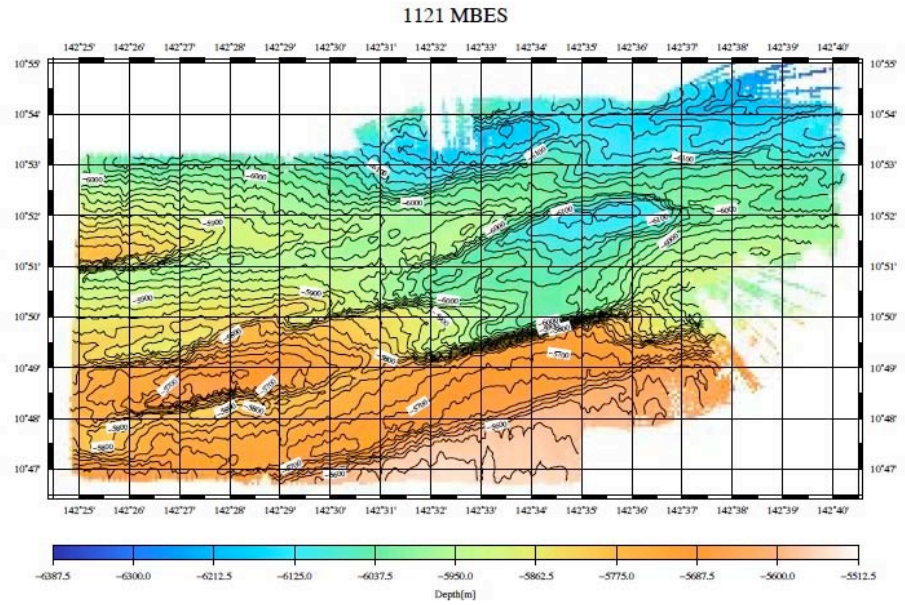
Kazumasa Oguri, Hiroshi Kitazato

Depth, temperature and salinity of water column were measured by SBE 49 CTD system mounted on the camera system. The CTD recorded pressure, conductivity and temperature from release to the recovery of the camera system. The depth was obtained to convert the pressure. Deployment date and the depth are shown in table 6-1.

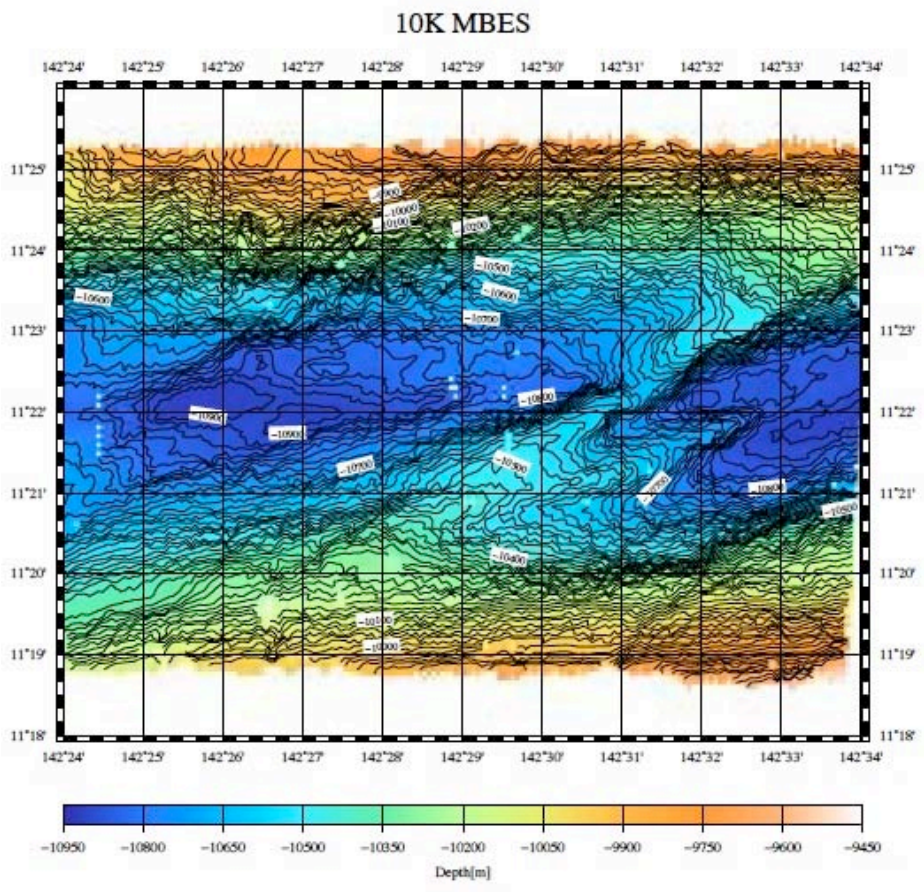
Table 6-1: Deployment date and the depth obtained by the CTD.

	Date	Maximum CTD depth (m)
Camera 1	2010/11/21	6037
Camera 2	2010/11/22	6038
Camera 3	2010/11/23-24	No data (failure)
Camera 4	2010/11/25-26	10900
Camera 5	2010/11/27-28	10900

2) Seabeam Map



Seabeam map of 6000m site



Seabeam map of 10000m site

3) Captured video

Kazumasa Oguri, Hiroshi Kitazato

Sediment surface and organisms were recorded by HDTV camera of the camera system. The camera was set to record sediment surface. Two deep sea lights were set to the direction for the recording images. The video recording was attempted for five times. Without fifth deployment (Camera 5), the recordings were succeeded. Detail of the video recording are shown in the table. Typical images of the sea floor and organisms in each deployment are shown in the figure.

Table6-2. Deployment of the video recording.

	Date	Maximum CTD depth	Baited trap	Video record
Camera 1	2010/11/21	6037		
Camera 2	2010/11/22	6038	×	
Camera 3	2010/11/23-24	No data (failure)		
Camera 4	2010/11/25-26	10900	×	
Camera 5	2010/11/27-28	10900	×	×

Figure 6-1. Video captured images recorded in Camera 1 at 6037m. Amphipods, red shrimps, unknown crustacea, polychaeta? tube, and fish are observed.

Figure 6-1 (continued). Video captured images recorded in Camera 2 at 6038m. Without baited trap, little organisms are observed.

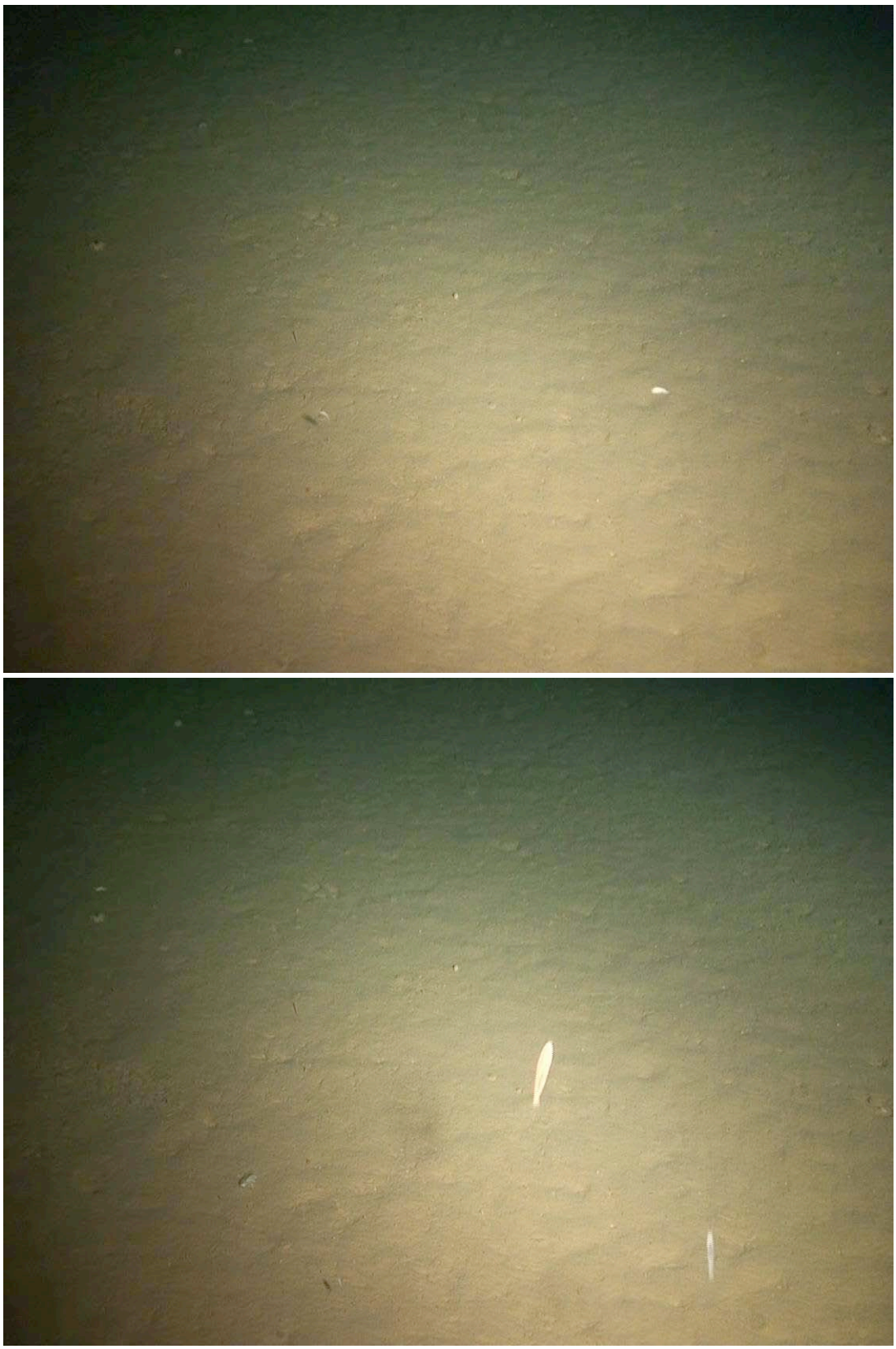


Figure 6-1 (continued). Video captured images recorded in Camera 3 at 10900m. Amphipods and benthic organism are observed. Sediment surface has weak

ripple-like structure.

Figure 6-1 (continued). Video captured images recorded in Camera 4 at 10900m. Tube

made by polychaeta? is seen at the sediment surface. Without baited trap, very little organisms are observed.

7. Cruise Summary

Five deployments of camera system were carried out during the cruise. 15 cores, video records and 140 scavenging amphipods specimens were obtained in the baited trap at the Challenger Deep area. Among 15 cores, 6 cores were processed for foraminiferal faunal analyses, specific level foram DNA analyses and environmental DNA analyses. One core was also processed for environmental DNA analyses. Six cores were used for on board oxygen / nitrogen profiles. Microbiological study will be don with the cores. Some chemical analyses will be done for one core. ^{210}Pb analyses plan to be made with one core for calculating mass accumulation rate of sediments at the Challenger Deep.

Four deployments were operated for ultra-deep lander. O_2 profiles at sediment-water interface were gotten at both sites. In situ O_2 measurements at the Challenger Deep were the first attempt in the world. In total, 70 profiles were obtained from the Challenger Deep. 64 profiles were also obtained from the 6000m site.

General video images both at 6000m and 10850m sites show strong contrast between both sites. Shallower site shows much dense and active benthic organisms, such as fish, amphipods and polychaetes. In contrast, less dense benthic organisms such as amphipods, horothrian and polychaetes, are seen at deeper site. In general, they move very slowly at the Challenger Deep.

Generally speaking, free-fall type camera-lander system works very well for hadal deep researches. It can recover different data and samples. It should be developed for a good platform of researches on hadal depth.

8. Notice on using

This cruise report is a preliminary documentation as of the end of the cruise. It may not be corrected even if changes on content (i.e. taxonomic classification) are found after publication. It may also be changed without notice. Data on the cruise report may be raw or not processed. Please ask the Chief Scientist for the latest information before using.

Users of data or results of this cruise are requested to submit their results to Data Integration aned Anaylsis group (DIAG), JAMSTEC.