

Yokosuka Cruise Report

YK13-02

Biological, geochemical and geological expedition to the hydrothermal fields in the Central Indian Ridge



February 6, Port Louis – February 25, Port Louis, 2013

Japan Agency for Marine-Earth Science & Technology

(JAMSTEC)

Acknowledgements

We are grateful to Captain Mr. S. Ryouno, Chief Officer Mr. T. Adachi and Chief Engineer Mr. E. Sakaguchi for their safe navigation and their skillful handling of “R/V Yokosuka”. Great thanks are due to Submersible Operation Manager Mr. T. Sakurai and “SHINKAI6500” operation team for their operations in sampling. We also thank Mr. M. Ito, Nippon Marine Enterprise, Ltd., for his attentive supports. We thank all the JAMSTEC personnel who have supported us. Finally, we would like to appreciate all the person who supported directly or indirectly this cruise.

CONTENTS

Acknowledgements	2
Contents	3
Cruise information	4
List of participants	6
Scientific group	6
Marine Technician	7
Captain and crew of the R/V YOKOSUKA	8
“Shinkai 6500” Operation Team	8
I. Cruise summary	10
II. Introduction	12
1. General background and objectives	12
2. Objectives of the cruise	12
III. Explanatory note	16
1. Manned Research Submersible “SHINKAI6500”	16
2. Research Vessel “Yokosuka”	16
3. Geochemistry	17
4. Microbiology and macrobiology	19
IV. Dive report	20
#1324 DIVE (J. Miyazaki)	21
#1325 DIVE (S. Kawagucci)	24
#1326 DIVE (G. Beedessee)	27
#1327 DIVE (M. Nishizawa)	30
#1328 DIVE (B. Vishwakalyan)	33
#1329 DIVE (Leckraz S. Kumar)	36

Cruise information

Cruise ID: YK13-02

Vessel: YOKOSUKA

Title of the cruise: Biological, geochemical and geological expedition to the hydrothermal fields in the Central Indian Ridge

Title of the proposal:

IS12-01: Experimental study for sclerite formation in the scaly-foot gastropod based on onboard culture experiments (Proponent: Dr. Manabu Nishizawa, JAMSTEC)

IS12-02: Clarification of the diversity of geo- and bio-systems and of the potential principles linking the systems in Central Indian Ridge hydrothermal activities (Proponent: Dr. Ken Takai, JAMSTEC)

IS12-05: Understanding of developmental process of morphological changes associated with symbiotic acquisition in hydrothermal vent gastropods (Proponent: Dr. Hiromi Watanabe, JAMSTEC)

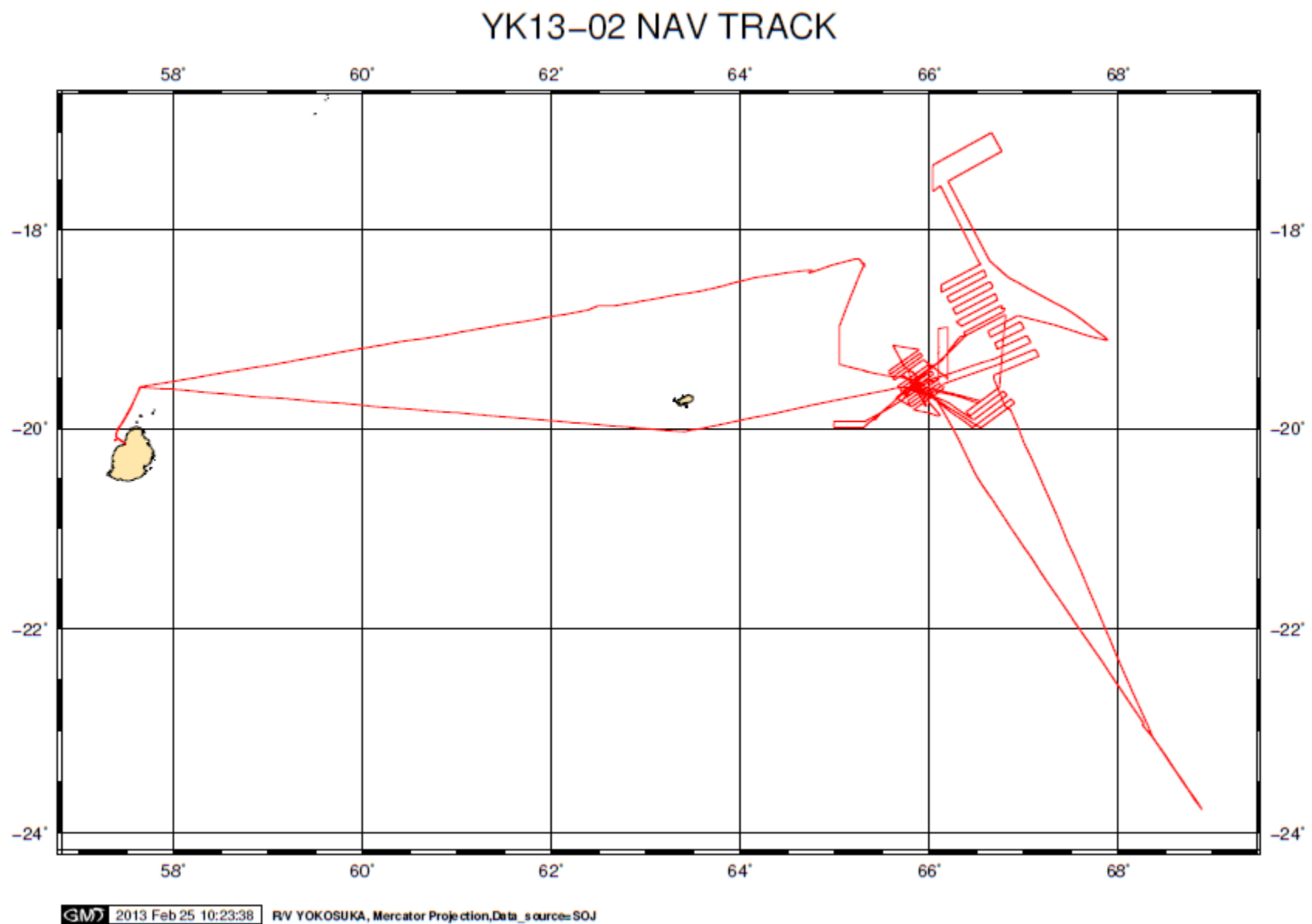
Cruise period: February 6 – February 25, 2013

Ports of call: Port Louis – Port Louis

Research area:

1. The Rodriguez Triple Junction in the Central Indian Ridge (CIR)
2. The CIR segments 15 and 16 at 18°-20°S

Research map:



List of Participant

Scientific party

Chief Scientist

Dr. Manabu Nishizawa

Research Scientist (Biogeochemistry)

Precambrian Ecosystem Laboratory (PEL)

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

Vice Chief Scientist

Dr. Ken Takai

Program Director (Geomicrobiology)

Subsurface Geobiology Advanced Research (SUGAR) Program & Precambrian Ecosystem Laboratory

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

Staff Scientists

Dr. Hiromi WATANABE

Research Scientist (Biology)

Marine Biodiversity Research Program, Institute of Biogeoscience

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

Dr. Junichi Miyazaki

Research Scientist (Microbiology)

SUGAR Program & Precambrian Ecosystem Laboratory

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

Dr. Tomoo Watsuji

Research Scientist (Biology)

SUGAR Program

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

Dr. Shinsuke Kawagucci

Research Scientist (Geochemistry)

SUGAR Program & Precambrian Ecosystem Laboratory

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

Dr. Kentaro Nakamura

Research Scientist (Alteration petrology)
Precambrian Ecosystem Laboratory
Japan Agency for Marine-Earth Science & Technology (JAMSTEC)

Dr. Ryuji Kondo

Associate Professor (Biology)
Department of Marine Bioscience
Fukui Prefectural University

Ms. Sayaka Mino

Ph. D student (Biology)
Laboratory of Microbiology, Faculty of Fisheries Sciences, Hokkaido University

Mr. Vishwakalyan BHOYROO

Research Scientist (Biology)
Mauritius Oceanography Institute (MOI)

Mr. Girish BEEDESSEE

Research Scientist (Biology)
Mauritius Oceanography Institute (MOI)

Dr. Prithivi Dass BISSESSUR

Research Scientist (Geophysics)
Mauritius Oceanography Institute (MOI)

Ms. Modoosoodun KHISHMA

Research Scientist (Biology)
Mauritius Oceanography Institute (MOI)

Mr. Leckraz Sanjeev Kumar

Research Scientist (Biology)
Ministry of Agro-Industry, Food Production & Security (Fisheries Division)

Marine Technician

Mr. Masashi Ito

Marine Science Department, Nippon Marine Enterprises, LTD.

Captain and crew of the R/V YOKOSUKA

Captain	Shinya Ryouno
Chief Officer	Tatsuo Adachi
2nd Officer	Takeshi Egashira
3rd Officer	Syunsuke Fujii
Chief Engineer	Eiji Sakaguchi
1st Engineer	Takashi Ota
2nd Engineer	Kenta Ikeguchi
3rd Engineer	Katsuto Yamaguchi
Chief Electronic Operator	Takehito Hattori
2nd Electronic Operator	Syunsuke Fukagawa
3rd Electronic Operator	Yoshikazu Kuramoto
Boat Swain	Kazuo Abe
Able Seaman	Masanori Ohata
Able Seaman	Kaito Murata
Able Seaman	Takuya Miyashita
Sailor	Shinsuke Uzuki
Sailor	Ryoma Tamkura
Sailor	Kenta Nasu
No.1 Oiler	Kozo Miura
Oiler	Keita Funato
Oiler	Yuji Higashikawa
Assistant Oiler	Makoto Kozaki
Assistant Oiler	Eiji Aratake
Chief Steward	Ryuei Takemura
Steward	Yoshio Okada
Steward	Seiji Honda
Steward	Kazuma Sonoda
Steward	Kei Ito

Operation team of SHINKAI 6500

Submersible Operation Manager	Toshiaki Sakurai
Deputy Submersible Op. Manager	Satoshi Ogura
Deputy Submersible Op. Manager	Kazuhiro Chiba
1st Submersible Technical Officer	Keita Matsumoto
1st Submersible Technical Officer	Masanobu Yanagitani

2nd Submersible Technical Officer	Hirofumi Ueki
2nd Submersible Technical Officer	Keigo Suzuki
2nd Submersible Technical Officer	Akihisa Ishikawa
2nd Submersible Technical Officer	Masaya Katagiri
3rd Submersible Technical Officer	Hitomi Ikeda
3rd Submersible Technical Officer	Yudai Tayama

I. CRUISE SUMMARY

Main scientific purposes of YK13-02 are to understand 1) geochemical and geobiological features of the recently discovered hydrothermal fields in the Indian Ocean (the DODO and Solitaire fields), 2) novel mechanism of sulfide biomineralization by scaly-foot gastropod, and 3) acquisition mechanism of end- and ecto-symbiosis by scaly-foot gastropod and *Alviniconcha* sp. In this cruise, we have successfully conducted six Shinkai6500 dives; one dive in the DODO field and five dives in the Solitaire field. Though we couldn't conduct Shinkai6500 dive at the Kairei and Edmond fields due to bad weather condition, we alternatively performed geophysical surveys, including multibeam bathymetry, magnetics and gravity surveys in the Rodriguez segment area (e.g. around the Solitaire and DODO fields and median ridge). The main results in this cruise are as follows:

- 1) Macrofaunal community structure in the Solitaire field was fully characterized by detailed observation and animal sampling. The macrofaunal structure in the Solitaire field, together with published data from the Kairei and Edmond fields (Hashimoto et al., 2001; Van Dover et al., 2001; Nakamura et al., 2012), will advance greatly our understanding of the biodiversity and biogeography of hydrothermal ecosystem in the Indian Ocean.
- 2) Physicochemical conditions of habitats of various hydrothermal vent animals (*Crysmallon squamiferum*, *Alviniconcha*, *Bathymodiolus*, *Phymorhynchus* and *Austinograea* spp.) in the Solitaire field were characterized by means of in-situ measurements of temperature and DO concentration of their colonies. Further information of their habitats will be obtained from shore-base analysis of water chemistry collected just above their colonies.
- 3) We successfully collected *Crysmallon squamiferum*, *Alviniconcha* and *Rimicaris* spp. for in-situ fixation of RNA. Those samples will provide important insights into their ecophysiology and/or interaction between host-animal and symbiotic microorganism.
- 4) Onboard cultivation experiments for *Crysmallon squamiferum* were successfully conducted in media with sulfur substrates labeled with ^{34}S or ^{35}S , and properly processed and stored. Isotope imaging of these samples will provide novel insights into the sulfide biomineralization.
- 5) Many hydrothermal fluid and chimney samples were obtained, and in-situ colonization system deployed at the Touon-3 chimney for 13 days were successfully retrieved. These samples would be enough for future geochemical, stable isotopic and geomicrobiological characterization of the Solitaire hydrothermal field.
- 6) Dive #1325 shows no signs of presently active hydrothermal fluid discharge in the DODO field, suggesting that hydrothermal activity in the DODO field had stopped during the period from 2009 to 2012.
- 7) The geophysical survey that was carried out during the YK13-02 expedition includes:
 - i. Bathymetry survey using the ship multibeam echo sounder (MBES), SeaBeam 2112
The purpose of the bathymetry survey was first to map the zones that have not been

mapped during previous expedition (fill the gaps) in order to obtain a complete bathymetry mapping of the Rodrigues segment area, and second to identify the structures of the seafloor that are difficult to identify on the satellite “predicted” bathymetry. Bathymetry profiles covering the gaps were carried out within the survey area (65° E - 68° E , 15° S - 20° S).

- ii. Magnetic survey using 1) a surface-towed magnetometer using a proton precession magnetometer, and 2) a 3-axis magnetometer installed on the ship that record the 3 components of the magnetic field.

The purpose of the magnetic survey was to record the magnetic signal of the oceanic crust in order to later investigate the age of the seafloor surrounding the Rodrigues segment of the Central Indian Ridge and its evolution. A dense magnetic survey of the Solitaire site was also carried out in the view of characterising the signature of a hydrothermal site.

- iii. Gravity data was acquired all along the ship tracks using the ship onboard gravimeter.

Gravity data will serve in the study the topography of the seafloor and the geodynamics of the area.

II. INTRODUCTION

1. General backgrounds

Since the first discovery of a hydrothermal vent site at the Galapagos Spreading Centre in 1977 (Corliss et al., 1979), submarine hydrothermal systems have attracted the particular interest not only of geoscientists, but also of chemists and biologists (Humphris et al., 1995; Van Dover, 2000; Wilcock et al., 2004). It is generally thought that submarine hydrothermal activities in the global ocean play a major role in elemental dynamics and exchange of vent-endemic unique animal communities. Even today, however, only 10% of the global mid ocean ridge system has been explored for hydrothermal activity.

The Indian Ocean is one of the most important target for mid ocean ridge hydrothermal system, along with the Pacific and Atlantic Oceans. A significant hydrothermal influx to the Indian Ocean has long been suggested by chemical signatures in sediments and seawater (Srinivasan et al., 2004). However, only five hydrothermal vent sites have been discovered in the Indian Ocean. Moreover, the Indian Ocean hydrothermal ecosystem is believed to represent a quite novel biogeographic province in the world (Van Dover et al., 2001, Hashimoto et al., 2001). In particular, a so-called ‘scaly-foot’ gastropod that has been discovered in the Indian hydrothermal vent sites has received broad attention of many researchers and public peoples due to its unique dermal sclerites coated by iron sulfide as armor (Warén et al., 2003; Suzuki et al., 2006). Additionally, a new type of scaly-foot gastropod devoid of iron sulfide coating was recently identified from a newly-discovered hydrothermal vent site in the Indian Ocean (Nakamura et al., 2012). Consequently, detailed understanding of the ecophysiology and sulfide biomineralization of scaly-foot gastropod is highly desired. Further, exploration of the unseen Indian hydrothermal activities, geochemistry and biodiversity promises great advances to understand geochemical cycle and biodiversity in the global ocean.

2. Objectives of the cruise

IS12-01: Experimental study for sclerite formation in the scaly-foot gastropod based on onboard culture experiments

The primary objective of IS12-01 is to understand sulfide mineralization process in *Crysmallon squamiferum* through onboard cultivation experiments. *Crysmallon squamiferum* was a hydrothermal vent-endemic gastropod first discovered from the Kairei hydrothermal field of the Central Indian Ridge (Warén et al., 2003). *Crysmallon squamiferum* was often called the scaly-foot gastropod due to the unique scale-shaped structures (called sclerites) coated by iron sulfides. No other organisms are known to produce a skeleton consisting of iron sulfides. Further, chemical and physical characterization of the sclerite suggests that iron sulfides (greigite, pyrite, mackinawite) form unique crystal structures with potential industrial applications (Suzuki et al., 2006). Despite particular importance in biology and material science, the detailed mechanism of sulfide mineralization in the sclerite was still unknown due to the lack of in-situ observation of sulfide

formation in the living individual. In the last research cruise in the Kairei hydrothermal field (YK9-13), the scaly-foot gastropod was, for the first time, successfully cultivated onboard over three weeks (JAMSTEC Press Release, 2009). Further, a new type of scaly-foot gastropod devoid of iron sulfide coating (“white scaly-foot gastropod”) was recently identified from the Solitaire field in the CIR (Nakamura et al., 2012).

In this cruise, we will conduct onboard cultivation experiments of the black and white scaly-foot gastropods in media with ^{34}S - or ^{35}S -labeled substrates. A series of the experiments will allow us to identify the sulfur substrate used in the sulfide mineralization (sulfate vs. hydrogen sulfide), the pathway of the substrate transport, and the exact place of sulfide mineralization in the sclerite.

IS12-05: Understanding of developmental process of morphological changes associated with symbiotic acquisition in hydrothermal vent gastropods

Most of benthic marine animals experience drastic morphological changes (e.g. metamorphosis and molting) in their life, and their traits are often changed associated with morphological changes, e.g. from planktonic larval life to benthic adult lice. In deep-sea hydrothermal vent, some animals harbor symbiotic bacteria inside or outside of their cells. These animals are usually larger than their relatives without symbionts, although both of them are inhabited in vent fields. The prominent examples must be scaly-foot gastropods in the family Peltospiridae and *Alviniconcha* in the family Provannidae. Most of peltospirids and provannids are about 1 cm in shell length, while scaly-foot and *Alviniconcha* gastropods are about 3 cm in shell length. In the case of *Alviniconcha*, the shell morphology of juvenile showed that they experienced more than two drastic morphological changes, and the latest one showed that their shell morphology was changed from typical provaniid one to *Alviniconcha* specific broad one. We hypothesized that this drastic morphological change may be accompanied with symbiotic acquisition, as both of them are required to specific characters of *Alviniconcha* among provaniid gastropods. With the scaly-foot gastropods which without any developmental morphology, we will describe developmental morphological changes of shell morphology and symbiont-harboring organs such as gills, esophageal gland, and sclerites, using morphological, developmental and microbiological techniques.

IS12-02: Clarification of the diversity of geo- and bio-systems and of the potential principles linking the systems in Central Indian Ridge hydrothermal activities

A primary scientific goal of IS12-02 is to understand the geochemical and biogeographical diversity of deep-sea hydrothermal activity in the world’s least explored ocean, Indian Ocean. The detail objective is to re-visit the recently discovered two hydrothermal fields on the Central Indian Ridge in the Mauritanian EEZ, the Dodo and Solitaire hydrothermal fields, by means of the manned research submersible ‘SHINKAI6500’, and to characterize the geological settings, the hydrothermal fluid chemistry and the associating microbial and macrofaunal communities throughout the seafloor observation, the onboard measurements and experiments of the shore-base analyses and experiments.

In September–November 2009, the first seafloor exploration of deep-sea hydrothermal activities in the Central Indian Ridge (CIR) at 18°–20°S was conducted using the SHINKAI6500 by an international research team of Japanese and Mauritian scientists. This expedition successfully witnessed the 3rd and 4th deep-sea hydrothermal fields, the Dodo and Solitaire hydrothermal fields, in the Indian Ocean. The discovery was quite significant and several new geochemical and biogeographical findings have been obtained from the observation, the onboard and the subsequent shore-base measurements and experiments. However, the seafloor observation was quite limited (totally 3 dives). In addition, the samples recovered from the seafloor, the geochemical analyses of hydrothermal fluids and the compositional and functional characterization of the microbial and macrofaunal communities are still insufficient to shed lights on the geological settings, the hydrothermal fluid chemistry and the associating microbial and macrofaunal communities of the new hydrothermal fields.

Thus, here, we, Japanese and Mauritian scientific team, are planning to re-visit the Dodo and Solitaire hydrothermal fields and to characterize the geological settings, the hydrothermal fluid chemistry and the associating microbial and macrofaunal communities throughout the seafloor observation.

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III. EXPLANATORY NOTE

1. Manned Research Submersible “SHINKAI6500”

Mission of “SHINKAI6500”

“SHINKAI6500” is able to operate surveys and observations down to the depth 6500 meters with one scientist and two pilots. During the operation, “SHINKAI6500” finds her position by two ways; Long Base Line system (LBL) and Super Short Base Line system (SSBL). The LBL system needs three bottom-mounted transponders to be deployed in the survey area. “SHINKAI6500” locates her own position by herself in real time and the mother ship determines the position of “SHINKAI6500” based on the position of transponders. The SSBL system does not require any transponder but the accuracy of the position is inferior to LBL system and “SHINKAI6500” cannot determine her own position.

Specifications

Length:	9.5m
Width:	2.7m
Height:	3.2m
Weight in air:	25.8t
Maximum operation depth:	6500m
Complement:	3 (2 pilots and 1 researcher)
Inner radius of pressure vessel:	2.0m
Normal dive time:	8 hours
Life support duration:	129 hours
Payload:	150kg (weight in air)
Under water speed:	0-2.0 knots (Emergency: 2.5 knots)
Observation instruments:	Pan-tilt-zoom color video camera Fixed-view color video camera 35mm still camera CTD sensors Gamma ray spectrometer CTFM sonar Video-image transmission system
Operating devices:	2 manipulators 2 retractable baskets

2. Research Vessel “Yokosuka”

Mission of “Yokosuka”

- 1) Operate submersible “SHINKAI6500”
- 2) Operate underway-geophysical equipments;

Multi Narrow Beam Echo Sounder (Sea Beam 2112.04)
 Gravity meter (Type S-63)
 Ship-borne three-components magnetometer (Type SFG-1212)
 Proton magnetometer (Typ STC10)

Research Facilities

In wet laboratory, a fumigation chamber, Milli-Q water purification system, -80°C and -20°C freezer, incubator and rock saw are equipped. In addition, “Yokosuka” has on-board video editing system for DVCAM, S-VHS and VHS.

Specifications

Length:	105.22m
Breadth:	16.0m
Height:	7.3m
Draft:	4.5m
Gross tonnage:	4439t
Cruising speed:	about 16kts
Cruising range:	about 9000mile

3. Geochemistry

WHATS fluid sampler

WHATS-II (Water Hydrothermal Atsuryoku Tight Sampler II) was developed for collecting hydrothermal fluid samples without any loss and atmospheric contamination of gas species. Since it overflows its sample bottle with sampling fluids, it is rather easy to collect fluids close to the endmember. This sampling system consists of inlet tubing, 4 pressure-resistant sample bottles with ball valves at both ends (volume of one bottle: 150 ml), an arm to open and shut the valve and a deep-sea compatible pump. Usually WHATS is installed just below the shell of Shinkai 6500 and a sample inlet is handled with a manipulator (Fig. 3-8). Operation is controlled from inside the shell. At the time of each sampling, fluid temperature can be monitored using a thermometer attached to the top of the inlet tube. It takes about 7 minutes to fill up one sample bottle of 150 ml capacity. Detailed description of the system is shown bellow.

Description

Dimension of frame:	600 mm × 660 mm
Weight:	35.2 kg in air 28.0 kg in seawater
Depth range:	4000 m
Sample volume:	150 ml × 4

Sampling rate: 75-300 ml / min
Electricity: DC24 V / 1.0 A

Treatment and onboard analyses of WHATS samples for gas chemistry

In general, for vent fluid sampling, the WHATS-II sample bottles were in pairs, with one of the bottles used for the analysis of soluble components chemistry and the other for gas chemistry. The bottle devoted to gas chemistry was processed on board using a high vacuum line specifically designed for this purpose using the following procedure. After the WHATS bottle is connected to the vacuum line, all of the connecting lines are evacuated to high vacuum using an oil diffusion pump. When sufficient vacuum is achieved, the vacuum line is closed in a static condition, and the valve on the bottle is opened and the water plus gas is allowed to drop into an evacuated flask. Sulfamic acid or cadmium chloride is added to the flask prior to the extraction in order to acidify the sample and aid in the extraction of carbon dioxide or to precipitate H₂S gas and dissolved sulfide in the hydrothermal fluid as CdS for the subsequent sulfur isotope analysis. The water in the extraction flask is then agitated by stirring bar. The gas phase was transferred to a total of 150 cm³ evacuated stainless steel and glass container (for 34 compositional and isotope measurements of gas components). After the gas phase was obtained, the liquid phase was taken into a 50 cm³ polypropylene bottle (for major cation and anion measurements). The obtained gas sample in the glass container with a butyl rubber stopper was balanced with ultrapure He gas and the gas pressure was monitored by a manometer. Then, gas components were quantified by GC-PID system on board.

Onboard analyses for dissolved species

The bottle devoted to fluid chemistry was shared with microbiological study. After sample for pH, Alkalinity, and H₂S determination was drawn, the rest of the fluid was filtered with a 0.2μm disk filter. The filtrate was provided for chemical analysis of major elements, nutrients (NH₄, SiO₂), and trace metals. The filtrate aliquot for trace metals was acidified with nitric acid to avoid hydroxide precipitation during storage. Because some chemical species such as H₂S and pH are difficult to be conserved during storage, we therefore analyzed these species onboard. In this cruise, colorimetric methods and titration were employed for onboard analyses as described below. These analytical methods are conventional ones and summarized in Gieskes et al. (1991).

pH and alkalinity

Determination of pH at room temperature was conducted with a pH meter with a combined glass electrode (Metrohm, 794 Basic Titrino). Measurements were done within an hour after sample distribution from the WHATS bottle. Calibration was conducted daily using JIS standard buffer solutions (pH=6.865 and 4.010). Alkalinity was determined by titration with hydrochloric acid. For calculation of the endpoint, Gran plot is employed using the pH/ion meter (Metrohm, 794 Basic

Titrimo).

Colorimetric method

Using a colorimeter (Shimazu, UV mini 1240), concentrations of hydrogen sulfide (H₂S) were analyzed with methylene blue method ($\lambda=670\text{nm}$). Analytical precision is usually better than 3% for seawater analysis, although sometimes the precision is somewhat worse for the case of hydrothermal fluids due to wide range of concentrations.

4. Microbiology and macrobiology

For cultivation, water samples collected by the Niskin bottle and WHATS were immediately poured into sterilized glass vials under the atmosphere of nitrogen gas. Chimney samples were subsampled into several portions (e.g. vent orifice surface, inside structure, middle-inside structure). Each piece of chimney structure was slurried with filter-sterilized seawater under N₂ for cultivation. For molecular analysis, the rest of pieces was kept under -80 °C. Hydrothermal vent animals were dissected, incubated, or fixed once onboard ship. Individuals or their tissues were applied to various onboard experiments, or kept under -80 °C. Some individuals of hydrothermal vent animals were frozen under -80 °C or fixed with ethanol or formalin.

References:

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IV. DIVE REPORTS

#1324 DIVE (DODO Field)

Dr. Junichi Miyazaki

#1325 DIVE (Solitaire Field)

Dr. Shinsuke Kawagucci

#1326 DIVE (Solitaire Field)

Mr. Girish Beedessee

#1327 DIVE (Solitaire Field)

Dr. Manabu Nishizawa

#1328 DIVE (Solitaire Field)

Mr. Bhoyroo Vishwakalyan

#1329 DIVE (Solitaire Field)

Mr. Leckraz Sanjeev Kumar

Dive Report: SHINKAI 6500 Dive#1324**Date:** February 8, 2013**Site:** DODO hydrothermal field in CIR segment 16**Landing:** 11:23; 18°20.2000'S, 65°18.0498'E, 2750 m**Leaving:** 15:54; 18°20.0253'S, 65°18.0820'E, 2740 m**Observer:** Junichi Miyazaki (JAMSTEC)**Pilot:** S. Ogura **Co-Pilot:** A. Ishikawa**Objectives:**

Objectives of this dive were collecting hydrothermal vent chimneys, vent fluids and animals in DODO hydrothermal field to characterize geochemical and biological features of DODO hydrothermal field. And also to capture microbes in hydrothermal fluids, we tried to deploy in situ colonization systems (Miyazaki11's short Bio sampler II) in the vent.

Dive Summary:

At 11:23, we landed on pillow lava which was located at south east side of previously found vent sites. After the position calibration, we head to vent sites in the 6k dive #1165 & 1166. At 12:14, we arrived at #99 marker which was set near Potsunen chimney. However, we could not observe discharging fluids. And we could not find 6k #100 marker deployed near Tsukushi-2 chimney which was located to only 10 m south position. But we could find the dead chimneys in the south area of the marker #99. In #99 site and the south site, we could not observe vent endemic faunas although we could find a brown crab. Therefore we concluded that hydrothermal activities in these sites had stopped during the period from 2009 to 2012.

To find active vent sites, we moved south, but we could not find any signs of hydrothermal activity. Therefore, when we arrived at 200 m south position of the marker #99 site, we headed west. After moving 80 m, we went north and headed to sites in which vent endemic faunas were observed in previous dives. However, no faunas were found. Therefore, we went back to the dead chimney site. After arriving, we sampled dead chimney.

We head to the landing point to find vent site. On the way, we found iron mats. Although we thought that these mats were active, shimmering was not observed. Again, we headed to landing point. But we could find neither active vent nor vent endemic faunas. At 15:54, we left the bottom.

Payloads:

- 1) Suction sampler
- 2) WHATS with 4 bottles
- 3) Cheap WHATS with 4 bottles
- 4) Miyazaki chimney box
- 5) 2 x Sample boxes

- 6) Miyazaki11's short Bio sampler II
- 7) 3 x Marker (#)

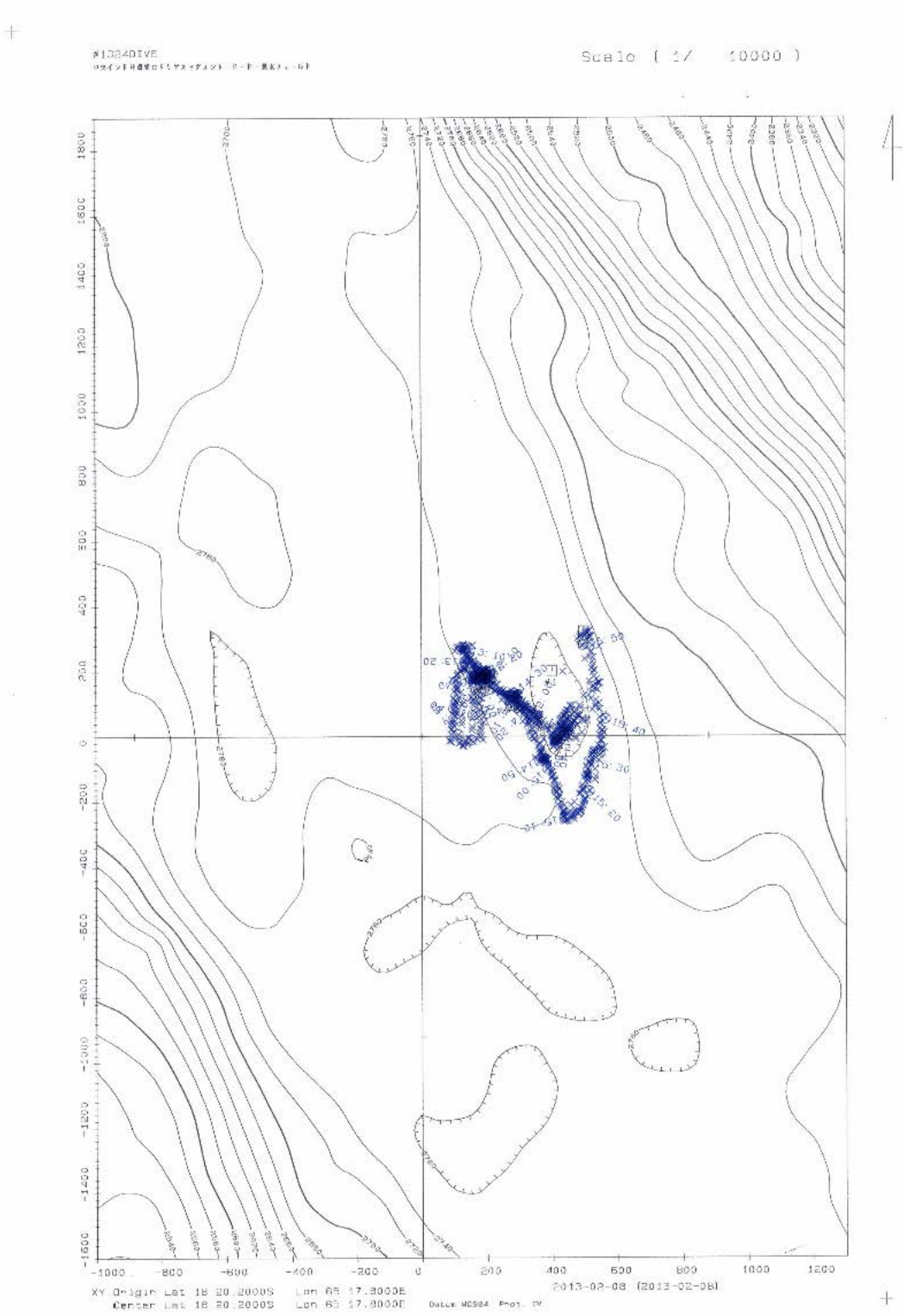
Event List:

- 1) 11:23, 18°20.2000'S, 65°18.0271'E, D=2742m Landing.
- 2) 12:14, 18°20.0954'S, 65°17.9190'E, D=2742m, Finding 6k marker #99.
- 3) 12:23, 18°20.1026'S, 65°17.9105'E, D=2743m, Finding dead chimneys
- 4) 13:56, 18°20.1046'S, 65°17.9041'E, D=2743m, Sampling dead chimney
- 5) 14:10, 18°20.1362'S, 65°17.9609'E, D=2742m, Finding iron mats
- 6) 15:54, 18°20.0253'S, 65°18.0820'E, D=2740m, Left bottom

Sample List

Sample ID	Samples	Site	Number
R-1	Dead chimney	18°20.1046'S, 65°17.9041'E, D=2743m	1

Dive Track:



Dive Report: Shinkai 6500 Dive#1325**Date:** February 9, 2013**Site:** Solitaire hydrothermal field (CIR segment 15B)**Landing:** 11:09; 19°33.4247'S, 65°50.8657'E, 2581 m**Leaving:** 15:33; 19°34.2367'S, 65°50.1318'E, 2319 m**Observer:** KAWAGUCCI, Shinsuke (JAMSTEC)**Pilot:** K. Chiba **Co-Pilot:** K. Suzuki**Objectives:**

The dive #1325 planned to take high-temperature hydrothermal fluid for understanding fluid geochemistry in the Solitaire field and its temporal change from 2009 to 2013. To collect benthic macrofauna, particularly “white scary-foot” gastropod, *Alviniconcha*, and *Rimicaris*, for onboard incubation experiments was another objective.

Dive Summary:

At 11:09, we landed on steep slope covered by crushed pillow lava at ~50 m SW and ~20 m above from DT#100 and 6K#1168 markers. In spite of some distance from the known active center, dense barnacle colony was identified. Then, Shinkai 6500 directed to the Tokon-3 chimney from SE passing through two markers. More than 20 patch-like colonies of the white scary-foot gastropod, each of which included >300 individuals, were found in each several meters in horizontal. Following to sampling high-temperature hydrothermal fluid ($T_{\max} = 302\text{ }^{\circ}\text{C}$) by WHATS (W1 - W4, but W2 was failed) until 12:40, the improved In-Situ Colonization System (ISCS; named Miyazaki11's short Bio sampler II) was penetrated into the mound at downward foot of the ~5m-height Tokon-3 chimney and the PIN of the ISCS was removed. We slightly moved south and approached to one of the colonies of white scary-foot gastropod. After measuring colony-water chemistry by DO sensor and a temperature probe on the fluid sampler ($T_{\max} = 38\text{ }^{\circ}\text{C}$) for 5 minutes, >10 and >50 individuals of the white scary-foot gastropods were taken by in-situ fixation and single canisters, respectively. With similar manner to the white scary-foot gastropods colony, >5 and >20 individuals of *Alviniconcha* sp. type-3 were taken by in-situ fixation and single canisters, respectively, after the environmental measurement ($T_{\max} = 33\text{ }^{\circ}\text{C}$). Low-temperature diffusive fluid (colony water) of the *Alviniconcha* sp. type-3 was sampled by 8-fold ORE-type fluid sampler for analyzing gas chemistry. We then moved to Tenkoji chimney, that emerged vigorous black smoker fluid, and took *Rimicaris kairei* and chimneys, but high-temperature fluid was not sampled at this dive. At last, we went to SW direction to discover another hydrothermal fluid venting site at Beauty Terrace, where several chimney structures had been suggested by geophysical survey by AUV r2D4 in KH-06-4 cruise. At Beauty Terrace, we identified the seafloor covered with white sediment on pillow lava. However, no hydrothermal signature (benthic animals, water turbidity, and brown/black sediment) had been found there.

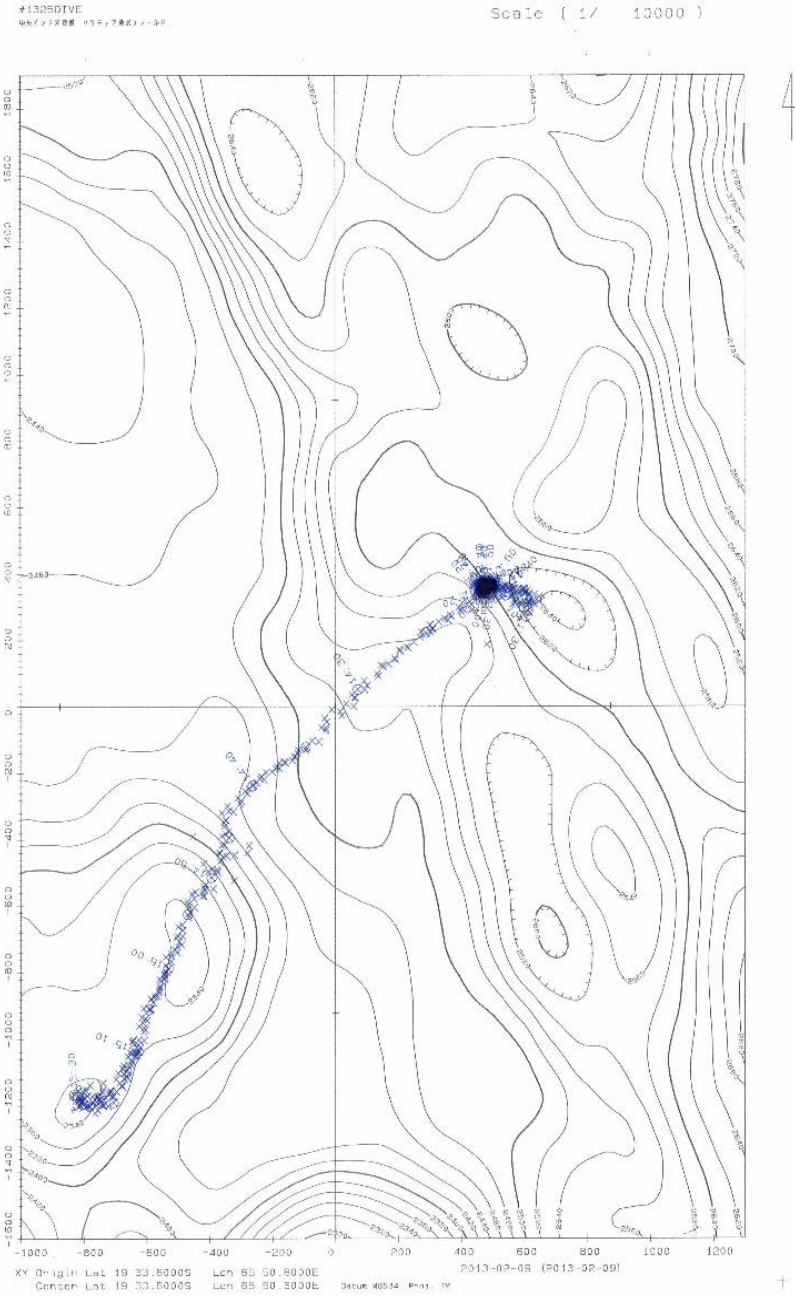
Payloads:

- 1) WHATS with 4 bottles
- 2) 8-fold ORE type fluid sampler
- 3) Suction sampler with a single canister
- 4) Suction sampler with a special canister for in-situ fixation by RNA later
- 5) Miyazaki11's short Bio sampler II
- 6) DO sensor
- 7) Markers (not used)

Event List:

- 1) 11:09, 19°33.4247'S, 65°50.8657'E, D=2581 m, Landing.
- 2) 12:40, 19°33.4063'S, 65°50.8802'E, D=2604 m, Tokon-3 chimney, Sampling WHATS (1-4)
T_{max} = 300 °C, ISCS deployment.
- 3) 13:26, 19°33.3991'S, 65°50.8714'E, D=2608 m, White Scary-foot colony, Measuring DO and T
and Sampling Scary-foot.
- 4) 13:40, 19°33.3991'S, 65°50.8718'E, D=2608m, *Alviniconcha* colony, Measuring DO and T.
Sampling *Alviniconcha* sp. and colony water.
- 5) 13:59, 19°33.4014'S, 65°50.8785'E, D=2608 m, Edge of Tenkoji chimney, Sampling *Rimicaris*
and chimney.
- 6) 15:33, 19°34.2367'S, 65°50.1318'E, D=2319 m, Beauty Terrace, Left bottom

Dive Track:



Dive report: Shinkai 6500 Dive#1326**Date:** February 10, 2013**Site:** Solitaire hydrothermal field (CIR segment 15B)**Landing:** 11:14; 19°33.3792'S, 65°50.9183'E, 2540 m**Leaving:** 15:40; 19°33.3823'S, 65°50.8886'E, 2395 m**Observer:** Girish Beedessee (Mauritius Oceanography Institute)**Pilot:** H. Ueki **Co-Pilot:** Y. Tayama**Objectives**

1. Collect vent fluid from Tenkoji chimney
2. Collect colony water by ORE-type sampler
3. DO measurement and collection of animals (Scaly Foot, *Alviniconcha*, mussels, crab and shrimps) from separate colonies and store in multi-bottled canister

Dive summary:

At 11:14, we landed north east of the Solitaire field and covered extended areas of pillow lava for nearly 30 minutes till we reached the site. Our first observation was the extensive population of phymorhynchus gastropods and barnacles far from any venting sites. Shinkai 6500 aligned itself with use of the two markers, DT#100 and 6k#1168 facing them in the initial stage of the dive.

White scaly-foot gastropods were distributed all around the venting site in small patches comprising of >100 individuals. We first collected colony water over a chosen colony of scaly-foot gastropods, followed by DO measurement for more than one minute of the same colony and finally collecting the same colony in the 6-fold canister.

We then performed DO measurement for nearly two minutes for phymorhynchus gastropods and collected the same in the 6-fold canister. Between 12:50 and 13:40 we took DO measurement for more than one minute for bathymodiolus mussels and alviniconcha gastropods in surrounding areas.

Around 13:00, we suffered from the damage of the handle of the DO sensor after which measurement of DO from colonies proved difficult. We made an attempt to take DO measurement from a colony of crabs but found it too risky to continue DO measurement. But we successfully collected bythograeidae crabs from one colony. We made an attempt to collect barnacles but without any success.

Our next objective was to collect chimney and WHATS sample. We didn't have enough time to search and approach Tenkoji chimneys and thus remained in the areas for chimney collection. We approached nearby chimneys and after much time spent to align Shinkai 6500, we were in good position to collect chimney structure near several grouped colonies of animals. Chimney displayed a maximum temperature of 85 °C and was collected for further investigation. Chimney fluid was collected by WHATS. The next plan was to observe and map the Solitaire field so as to better understand its setting but we lacked time for further investigation. We stopped the observation at

15:40.

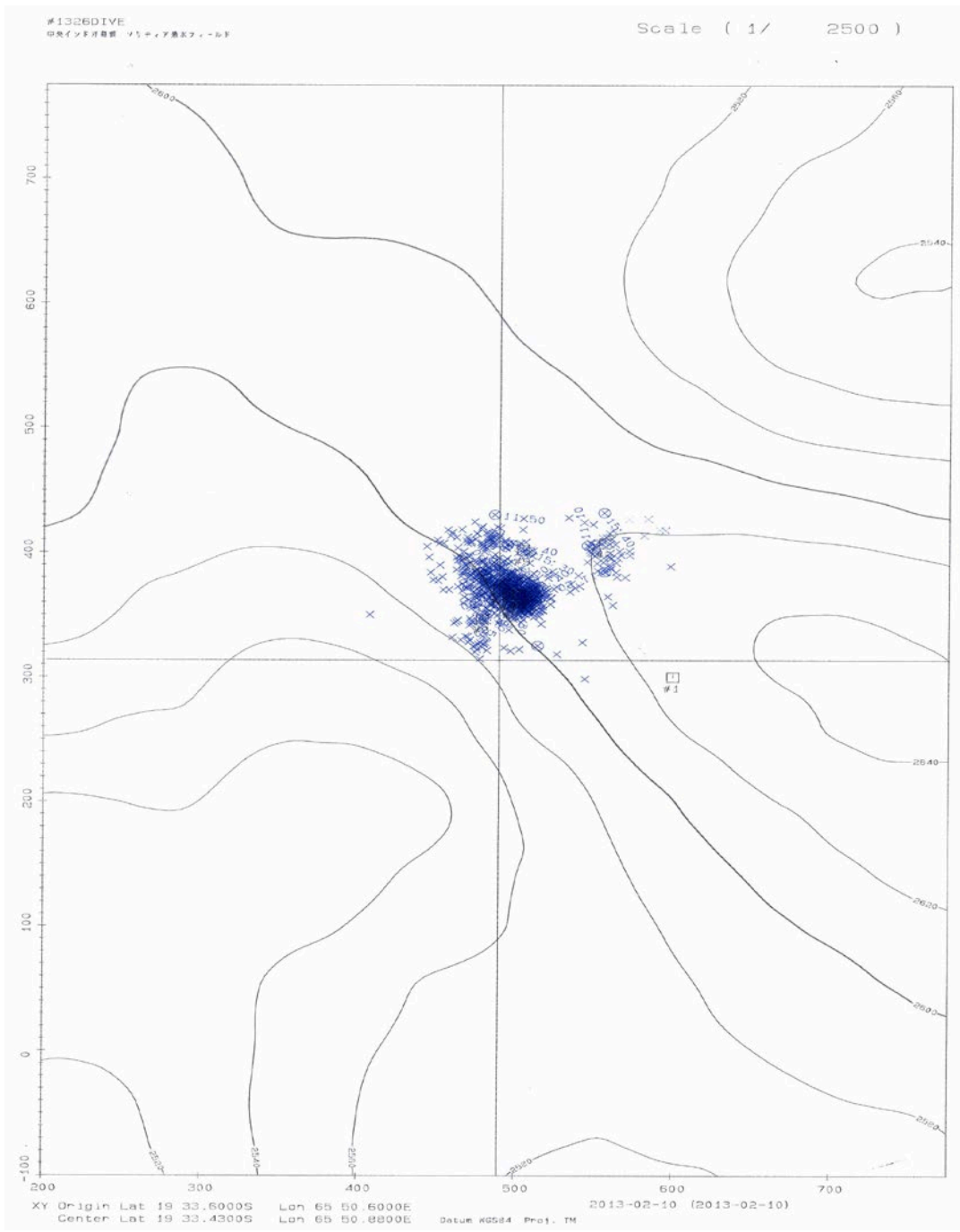
Payloads:

- 1) 8-fold ORE type fluid sampler
- 2) Multi-bottle (6-series) canister
- 3) Suction sampler
- 4) Sample box with lid
- 5) DO sensor
- 6) WHATS water sampler
- 7) Markers (not used)

Event List:

- 1.11:14 19°33.3792'S, 65°50.9183'E, D=2617 m, landing
- 2.12:10 19°33.4014'S, 65°50.8869'E, Sampling water Scaly foot gastropod; DO measurement and sampling scaly-foot gastropod and phymorhynchus, D=2608 m
- 3.13:09 19°33.4014'S, 65°50.8869'E, DO and Sampling bathymodiolus, D=2608 m
- 4.14:00 19°33.4054'S, 65°50.8844'E, DO and Sampling Alviniconcha, D = 2608 m
- 5.14:51 19°33.3991'S, 65°50.8800'E, Sampling Bythograeidae crab, D=2608 m
- 6.15:35 19°33.4025'S, 65°50.8834'E, Sampling chimney and WHATS, T= 85 °C, D=2607m
7. 15:40 19°33.3823'S, 65°50.8886'E, D=2607 m, Left bottom

Dive Track:



Dive Report: Shinkai 6500 Dive#1327**Date:** February 11, 2013**Site:** Solitaire hydrothermal field (CIR segment 15B)**Landing:** 11:15; 19°33.3375'S, 65°50.8890'E, 2596 m**Leaving:** 15:51; 19°33.4242'S, 65°50.8730'E, 2537 m**Observer:** Manabu Nishizawa (JAMSTEC)**Pilot:** K. Matsumoto **Co-Pilot:** M. Katagiri**Objectives:**

Main purposes of the dive #1327 were to collect a variety of benthic macrofaunal communities for understanding biogeography in the Solitaire Field, and to take high-temperature hydrothermal fluid venting from the Tenkoji chimney for understanding fluid geochemistry.

Dive Summary:

At 11:15, we landed on a slope covered by pillow lava at ~30 m north from the center of Solitaire field. Shinkai 6500 went southward to observe the unexplored northern area. At 11:28, we observed dead shapes of *Bathymodiolus* on a steep slope. Then we found sea cucumber, *Galatheidae* sp. and shellfish on the slope, and collected them in a multi-bottle canister (bottle 1).

At 12:01, we found a 1-m-height chimney (depth = 2621 m) on the slope. Maximum temperature of the fluids venting from the chimney was 90 °C. We observed colonies of *Alviniconcha*, *Phymorhynchus* and *Rimicaris* spp. near the chimney. We successfully measured the temperature and DO concentration of the water just above the colony of *Alviniconcha*, and then sampled *Alviniconcha* and *Rimicaris* spp. for in-situ fixation of RNA. Accidentally, we couldn't collect the water by 8-series water sampler. At 12:54, we sampled *Rimicaris* sp. in a multi-bottle canister (bottle 2). A marker #143 was deployed near the chimney.

At 13:45, we found a colony of white scaly-foot gastropod and measured the temperature and DO concentration of the water just above the colony. Then, we collected the scaly-foot gastropod and *Galatheidae* sp. for in-situ fixation of RNA. At 14:07, scale worm and shellfish around the colony of the scaly-foot were collected in a multi-bottle canister (box 3).

At 14:51, we collected apparently black-colored hydrothermal fluids (in two bottles of WHATS) venting from a chimney (depth = ~2610 m) near the Toukon-3 Chimney. Maximum temperature of the fluids was 306 °C. A marker #142 was deployed near the chimney (depth = 2608 m). Then, we tried to find the Tenkoji chimney, but couldn't find it.

At 15:21, we collected *Rimicaris* spp. in a multi-bottle canister (bottle 4). At 15:30, we collected rock samples with barnacle.

Payloads:

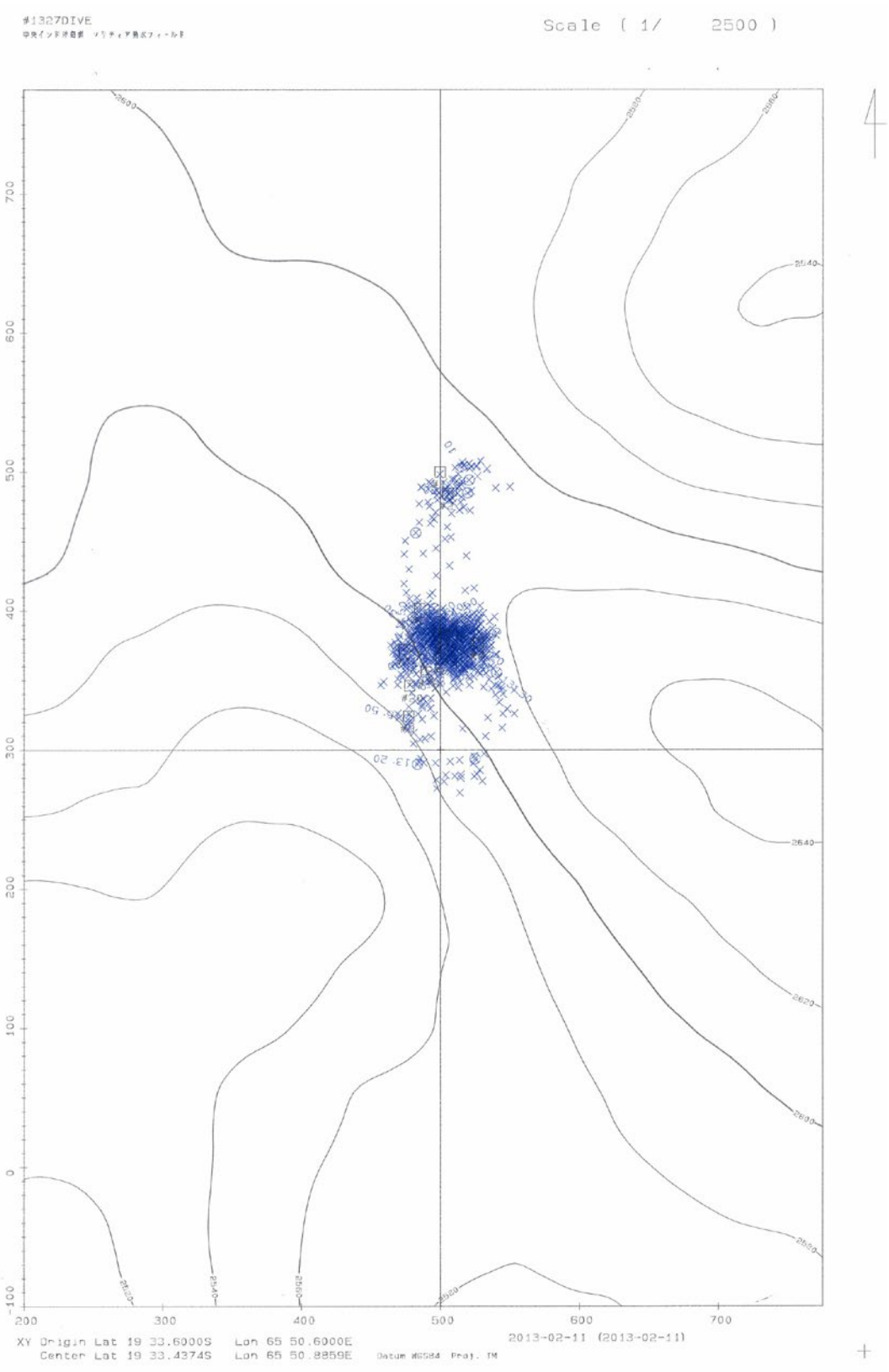
1) WHATS with 4 bottles

- 2) 8-fold ORE type fluid sampler
- 3) Suction sampler with a multi-bottle (6-series) canister
- 4) Suction sampler with a special canister for in-situ fixation by RNA later
- 5) DO sensor
- 6) Markers

Event List:

- 1) 11:15, 19°33.3375'S, 65°50.8890'E, D=2596 m, Landing
- 2) 11:57, 19°33.3982'S, 65°50.8711'E, D=2621 m, Sampling Sea cucumber, *Galatheidae*, Barnacle with rock
- 3) 12:50, 19°33.3956'S, 65°50.9016'E, D=2620 m, *Alviniconcha* colony, Measuring DO and T, Sampling fluids by 8-fold ORE type fluid sampler (failed)
- 4) 12:50, 19°33.3956'S, 65°50.9016'E, D=2620 m, *Alviniconcha* colony, Sampling *Alviniconcha* sp., *Rimicaris*, set #143 Marker
- 5) 14:14, 19°33.4018'S, 65°50.8863'E, D=2610 m, white scaly-foot colony, Measuring DO and T, Sampling scaly-foot.
- 6) 15:02, 19°33.3928'S, 65°50.8875'E, D=2608 m, apparently black smoker chimney, Sampling hydrothermal fluids by WHATS (two bottles), set #142 marker
- 7) 15:30, 19°33.3823'S, 65°50.8775'E, D=2604 m, Sampling *Rimicaris*, Rocks with barnacle
- 8) 15:51, 19°33.4242'S, 65°50.8730'E, D=2537 m, Left bottom

Dive Track:



Dive Report: Shinkai 6500 Dive# 1328**Date:** February 12, 2013**Site:** Solitaire Hydrothermal Field (CIR segment 15B)**Landing:** 11:06; 19° 33-6260'S, 65° 50.7727 E, D= 2506m**Leaving:** 14:26; 19° 33.3935'S, 65° 50.8753'E, D=2614m**Observer:** Bhoyroo Vishwakalyan (MOI)**Pilot:** M. Yanagitani **Co-Pilot:** H. Ikeda**Objectives:**

The dive #1328 planned to take high temperature hydrothermal fluid from the Tenkoji chimney for fluid geochemistry in the Solitaire field. To collect chimney samples from the Toukon Chimney for microbiological studies. To collect benthic macrofauna (Rimicaris, Alviniconcha and white-scaly foot) for onboard incubation experiments and also to collect planktons near active chimney, gastropod colony and barnacle colonies for larval studies.

Dive summary:

At 11.06, we landed near a sea mount with lava pillow at 2506 m deep. There was no sign of biological activity near the sea mount. Shinkai 6500 was then directed towards Tenkoji Chimney. At 11:38, we passed through the newly discovered chimney (MARKER 143). Tenkoji Chimney was reached at 11:44, and it was located north of the newly discovered chimney. Lots of barnacles, white gastropods, shrimps (Rimicaris and Choracharis), sea anemones and mussels were observed. Outside temperature at Tenkoji was around 1.3 °C. The temperature recorded at the vent ranged from 291 °C to 302 °C. High temperature hydrothermal fluid was collected in the WHATs sampler. Fluid sampling at Tenkoji Chimney finished at 12:40, following which Rimicaris was collected in the single canister near the chimney. After this, we moved to Toukon Chimney (2604 m deep), where we reached at 12:52. The outside temperature was 0.5 °C and lots of Rimicaris were observed on the chimney, and barnacles observed in the surrounding area. Chimney sample was collected in both the normal box and chimney fixation box. Sampling of chimney pieces at Toukon finished at 13: 26, following which Rimicaris was collected and we left at 13:50 from Toukon. We headed for biological sampling, when we were informed that we had only 30 minutes for biological sampling due to strong waves at sea surface. We prioritized sampling for white-scaly foot and Alviniconcha gastropods. We collected sample for white scaly foot (>15) gastropods which was very common, but we managed to collect only very few samples of Alviniconchia which was less common in the field. We left sea-floor at 14:26. At 1500m water was exchanged in the single canister.

Payloads:

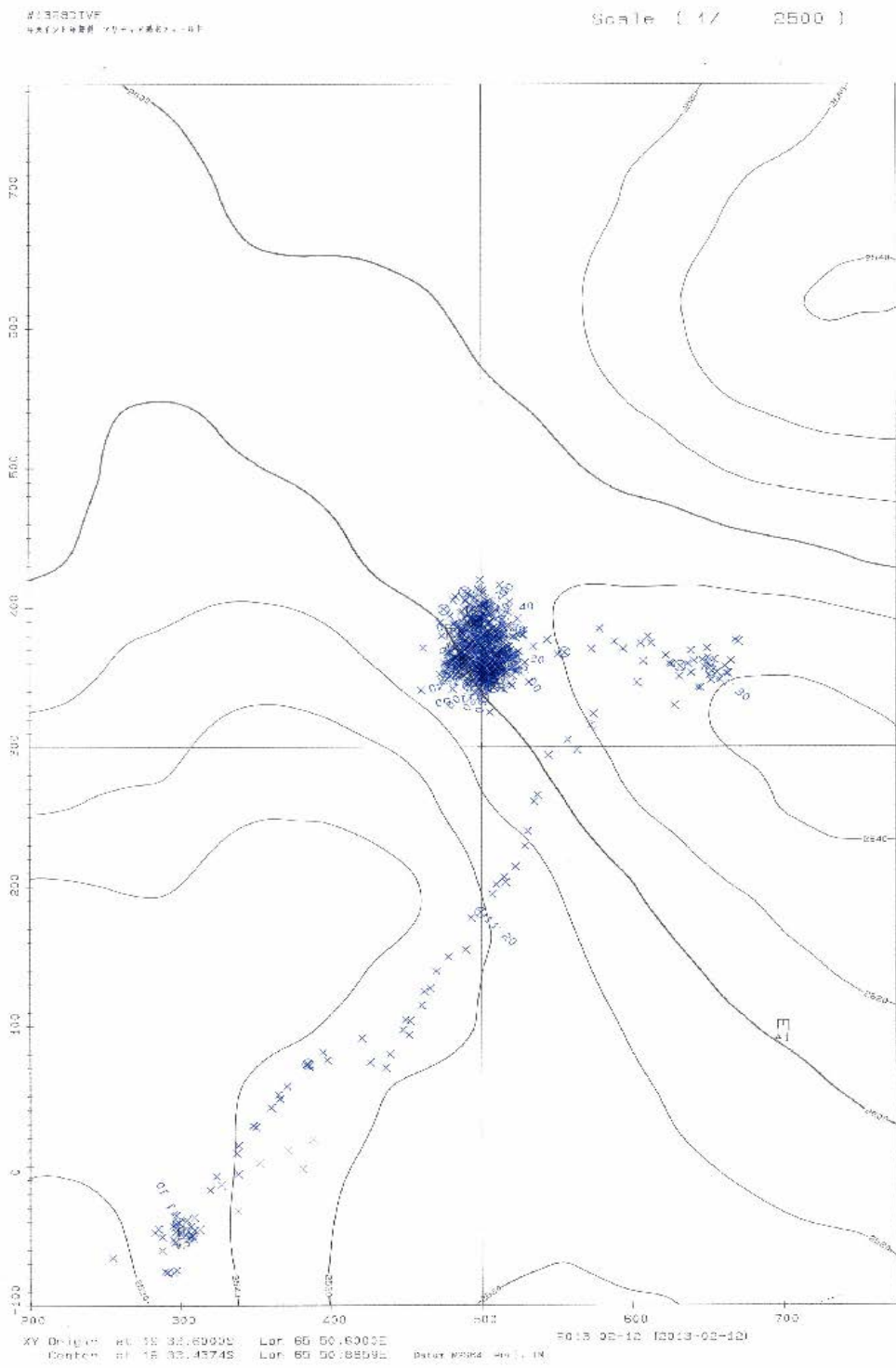
- 1) WHATs with 4 bottles

- 2) Miyazaki Chimney Box
- 3) Sample box (i.e. normal box) with lid
- 4) Single Canister sampler
- 5) Multi-bottle (6-series) canister

Event list:

- 1) 11:06, Sea bottom, 19° 33-6260'S, 65° 50.7727 E, D=2506m
- 2) 11:44, Tenkoji Chimney, 19° 33.4047'S 65° 50.8802'S, D=2608 m, Sampling WHATS (1-4)
Tmax=302 °C, Rimicaris in Single canister
- 3) 12:52, Toukon chimney, 19° 33.4090'S 65° 50.8924'E , D=2604m, Sampling Chimney in Normal
box and Fixation Box, Rimicaris in single canister
- 4) 14:24, biological sampling, 19° 33.3848'S 65°50.8827'E, D=2608m, White scaly foot and
Alviniconcha sampling.
- 5) 14:26, left bottom, 19° 33.3935'S 65° 50.8753'E, D=2614m

Dive Track:



Dive Report: Shinkai 6500 Dive# 1329**Date:** February 22, 2013**Site:** Solitaire Hydrothermal Field (CIR segment 15B)**Landing:** 11:19; 19° 33.3886'S, 65° 50.9643'E, D= 2619m**Leaving:** 15:26; 19° 33.4123'S, 65° 50.8651'E, D=2605m**Observer:** Leckraz Sanjeev Kumar (Ministry of Fisheries)**Pilot:** M. Yanagitani**Co-Pilot:** Y. Tayama**Objectives:**

The dive #1329 was planned to:

Collect water sample using 8 fold water sampler over *Alviniconcha* colony found near marker #143 chimney

Collect upper part of marker #143 chimney

Sampling low temperature fluid by WHATS sampler of marker #143 chimney

Measure DO over the *Alviniconcha* colony, white scaly foot colony, *Bathymodiolus* colony, *Phymorhynchus* colony and *Austinograea* colony

Collect *Alviniconcha* and white scaly foot individuals

Collect plankton above gastropods and barnacles colonies and near marker #143 chimney

Recover ISCS

Dive summary:

At 11:19, we landed on the sea bottom made up of lava pillow at a depth of 2619m. There was no sign of biological activity near the site we landed. Shinkai 6500 was then directed towards the Solitaire Field. At 11:30, we located the marker #143 where lots of barnacles, gastropods, shrimps (*Rimicaris* and *Chorocaris*), crabs (*Austinograea*), sea anemones and mussels were observed.

A colony of *Alviniconcha* (>30) was identified inside which DO measurement was carried out. Sampling of colony water was taken with the 8 fold water sampler. The water temperature varied between 6 to 31°C. *Alviniconcha* specimens were then collected in canister 2 using the multiple canisters. A colony of white scaly foot (WSF) was then identified. DO measurement (2 minute/points x3) followed by collection of specimens of WSF in canister 3 was done. A few *Phymorhynchus* specimens were also collected in canister 3.

We then collected the upper part of the Marker #143 chimney in Miyazaki's box. The maximum temperature recorded at the vent was 257 °C. Low temperature hydrothermal fluid was collected in the WHATS sampler. Planktons sampling over gastropods colonies was carried out in canister 4. After this exercise, Shinkai 6500 was directed towards the Tenkoji Chimney where another colony of white scaly foot was identified in order to carry out DO measurement (2 minute/points x5). DO measurements were also carried out above *Bathymodiolus*, *Phymorhynchus* and *Austinograea* colonies.

At about 14:55, Shinkai 6500 was directed towards the Toukon Chimney where the ISCS was recovered at 15:23. We left the sea-floor at 15:26. At 1500m water was exchanged in the multiple canisters.

NB: Planktons sampling near the Marker #143 chimney and over barnacles were not carried out due to some problems in the multiple canister.

Payloads:

- 1) WHATS with 4 bottles
- 2) 8-fold ORE type fluid sampler
- 3) Miyazaki Chimney Box
- 4) Multi-bottle (6-series) canister
- 5) DO sensor

Event list:

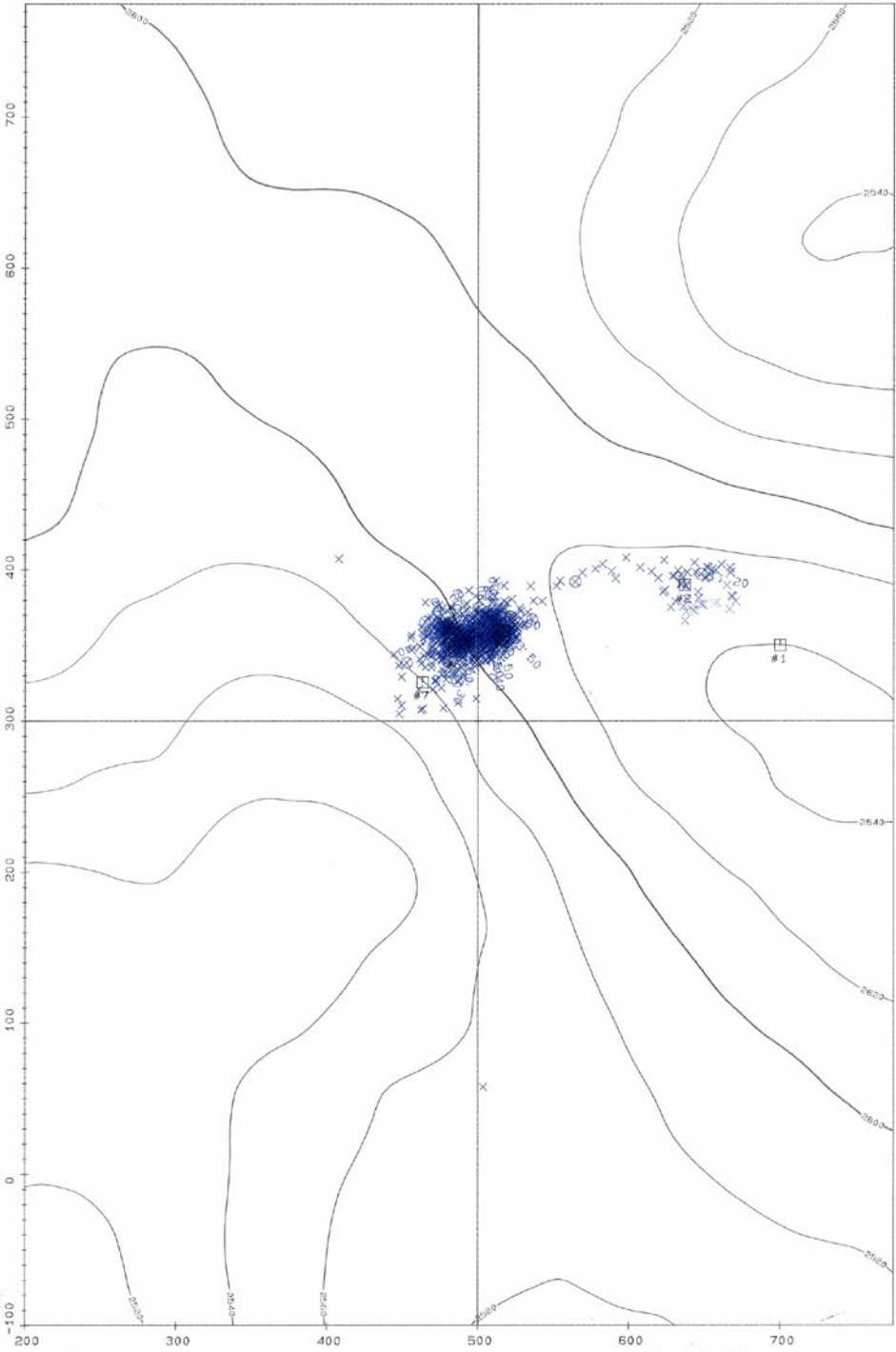
- 1) 11:19, Sea bottom; 19° 33.3886'S, 65° 50.9643'E, D= 2619m
- 2) 11:30, Marker #143 Chimney, 19° 33.4050'S, 65° 50.8956'E, D=2619m, DO measurement inside *Alviniconcha* colony (11:47:20 – 11:49:20), Water sampling by 8 fold sampler, Temp= 6 – 31 °C, *Alviniconcha* in Multiple Canister (Bottle 2), DO measurement inside WSF colony (2min/point x 3) – Pt 1 (12:23:30 – 12:25:30), Pt 2 (12:28:40 – 12:30:40), Pt3 (12:32:20 – 12:34:20), WSF in Mult. Canister (Bottle 3)
- 3) 12:44, Collection of chimney part in Miyazaki's box, Low temperature fluid sampling by WHATS Sampler (Tmax=257 °C)
- 4) 13:40, Sampling for planktons above gastropod colonies in Multiple Canister (Bottle 4)
- 5) 14:16, DO measurement inside WSF colony near Tenkoji Chimney (2min/point x 5), Pt 1 (14:18 – 14:20), Pt 2 (14:21:30 – 14:23:30), Pt3 (14:24:30 – 14:26:30, Pt 4 (14:33 – 14:35), Pt 5 (14:36 – 14:38), DO measurement above *Bathymodiolus* colony (14:30 – 14:32), *Phymorhynchus* colony (14:39:30 – 14:41:30) and *Austinograea* colony (14:46:30 – 14:48:30)
- 6) 14:55, Toukon3 Chimney, 19° 33.4123'S, 65° 50.8772'S, D=2602m
- 7) 15:23, Recovery of ISCS
- 8) 15:26, left bottom, 19° 33.4235'S, 65° 50.8651'E, D=2605 m

Dive Track:

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#1329DIVE
中央インテリジェンス マリチ、アモス、アモス、アモス

Scale (1/ 2500)



XY Origin Lat 19 33.6000S Lon 65 50.6000E 2013-02-22 (2013-02-22)
 Center Lat 19 33.4374S Lon 65 50.8859E Datum WGS84 Proj. TM

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