

RV Natsushima Cruise Report NT11-09

Hyper-Dolphin 3000 Dive Research

Myojin Knoll, Suiyo Seamount (Izu-Ogasawara Area) Off Hatsushima (Sagami Bay)

June15, 2011-June 26, 2011

Chief Scientist

Koji INOUE

(The University of Tokyo)

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

•Contents

1.	CRUISE INFORMATION	3
	1) Cruise ID/ Name of vessel	3
	2) Name of Vessel	3
	3) Title of cruise	3
	4) Title of proposals	3
	5) Cruise period	3
	6) Ports of call	3
	7) Research area	3
2.	RESEARCHERS	5
	1) Chief scientist	5
	2) Representative of the science party	5
	3) Member of the Science Party	5
3.	OBSERVATION	7
	1) Overview of the cruise	7
	2) Cruise Log	8
	3) Major equipments loaded to Hyper-Dolphin	10
	a) Slurp Gun (Suction sampler)	10
	b) Sample boxes	12
	c) Rotery clean seawater sampler (ROCS) and on-line thermometer	11
	d) Niskin water sampler, Van Dorn water sampler and MBARI corer	12
	e) Fish trap "Hodohodo-kun"	13
	4) Dive information	14
	a) Dive #1284	14
	b) Dive #1285	15
	c) Dive #1286	16
	d) Dive #1287	17
	e) Dive #1288	18
	f) Dive #1289	19
	g) Dive #1290	20
	h) Dive #1291	21
	i) Dive #1292	22
	j) Dive #1293	23
4.	RESEARCH INFORMATION (Methods and preliminary results)	24
	1) Studies on mechanisms of hypotaurine synthesis and its role in adaptation to toxic	24
	sulfide in hydrothermal vent-specific organisms	
	2) Culture experiment of Paralvinella hessleri in different tempgrature, and cDNA cloning of	24
	heat resistance relating gene.	
	3) Study on Symbiotic systems of deep-sea clam Calyptogena and intracellular sulfur	25
	oxidizing bacteria	
	4) Is it universal phenomenon for the hydrothermal vent community to detoxify hydrogen	26
	sulfide using thiotaurine? -Study on correlation between hydrogen sulfide concentration	
	and the amount of amino acids.—	
	5) Study on immunity in the deep-sea hydrothermal vent mussels	27
5.	ACKNOWLEDGMENTS	28
6.	NOTICE ON USING	28
Ар	pendix (point maps and track charts)	29
-		

1. CRUISE INFORMATION

- 1) Cruise ID: NT11-09
- 2) Name of vessel: R/V Natsushima
- 3) Title of the cruise: FY2011 Hyper-Dolphin 3000 Dive Research
- 4) Title of proposal
 - Studies on mechanisms of hypotraurine synthesis and its role in adaptation to sulfides in hydrothermal vent-specific organisms. (K. Inoue)
 - Culture experiment of *Paralvinella hessleri* in different tempgrature, and cDNA cloning of heat resistance relating gene. (T. Mori)
 - The analyses of symbiosis mechanism by in situ PLA technique and the symbiotic-specific monoclonal antibody library in connection with the *Calyptogena* symbiosis. (Y. Nakamura)
 - Is it universal phenomenon for the hydrothermal vent community to detoxify hydrogen sulfide using thiotaurine?: Study on correlation between hydrogen sulfide concentration and the amount of amino acids. (T. Koito)

Immune defense system of deep-sea hydrothermal-vent mussels belonging to *Bathymodiolus*. (K. Ohishi)

5) Cruise period

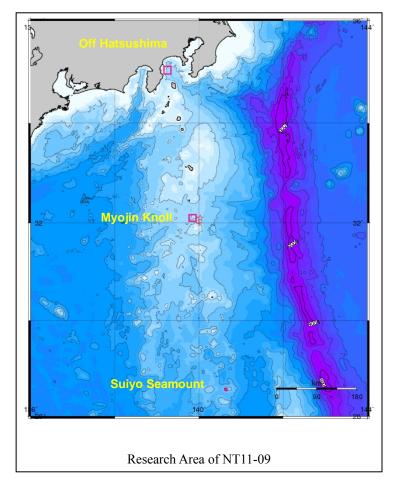
June 15, 2011-June 26, 2011

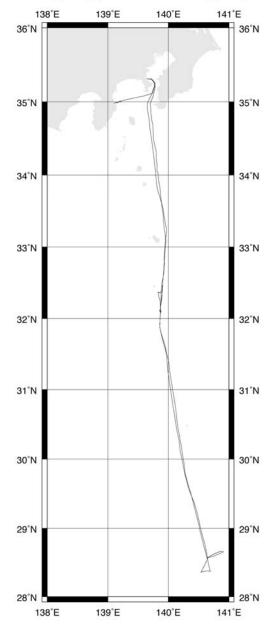
6) Ports of call

Yokosuka (JAMSTEC) to Yokosuka (JAMSTEC). (Change of Scientists on June 23 at Yokosuka)

7) Research area

Izu-Bonin Area (Myojin Knoll and Suiyo Seamount), Sagami Bay (Off Hatsushima)





NT11-09_R/V_NATSUSHIMA_ShipTrack

CMD 2011 Jun 25 22:24:16 NT11-09_RV_NATSUSHIMA_CruiseTime2011.6.15-2011.6.26_WGS84_MercatorProjection

Cruise track of NT11-09

2. RESEARCHERS

1) Chief scientist:

Koji INOUE [Atmosphere and Ocean Research Institute (AORI), The University of Tokyo]

2) Representative of the Science Party Koji INOUE [AORI, The University of Tokyo] Tsukasa MORI [Nihon University] Yoshimitsu NAKAMURA [JAMSTEC] Tomoko KOITO [Nihon University] Kazue OHISHI [JAMSTEC]

3)Member of the science party (on board) Ryusaku DEGUCHI (Miyagi Educational University) Koji INOUE (AORI, The University of Tokyo) Yuki HONGO (JAMSTEC) Azusa KINJO (AORI, The University of Tokyo) Madoka KITAJIMA (Enoshima Aquarium) Wen LIU (Kyoto University) Satomi MINAMIZAWA (Nippon Marine Enterprises, Ltd., Observation Technician) Yoshimitsu NAKAMURA (JAMSTEC) Toshihiro NAGASAKI (AORI, The University of Tokyo) Suguru NEMOTO (Enoshima Aquarium) Genki OZAWA (JAMSTEC) Kei SATO (The University of Tokyo) Daisuke SEKINE (JAMSTEC) Shuichi SHIGENO (JAMSTEC) Takuho SHUTO (AORI, The University of Tokyo) Makoto SUGIMURA (Enoshima Aquarium) Akihiro TAME (Marine Work Japan) Haruhiko TOYOHARA (Kyoto University) Takuya YAHAGI (AORI, The University of Tokyo) Shosei YAMAGAMI (Okayama University) Yu YAMAMOTO (AORI, The University of Tokyo) Takefumi YORISUE (AORI, The University of Tokyo) Takao YOSHIDA (JAMSTEC)



Science parties of NT11-09. First leg (upper) and second leg (lower).

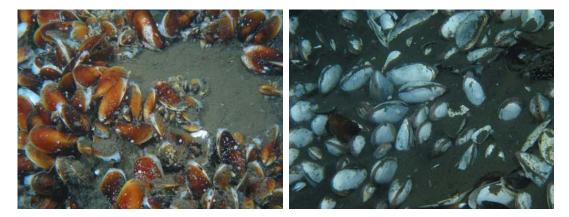
3. OBSERVATION

1) Overview of the cruise

In this cruise, two days of researches were performed at each of three areas, Myojin Knoll, Suiyo Seamount, and Off Hatsushima (6 days in total). Five research groups participated to this cruise. As the total number of scientists who wanted to join the cruise was more than the number of the bed of Natsushima, we divided the cruise into two legs; first one is to Izu-Ogasawara (Bonin) and the other one to Off Hatsushima. We replaced some members after the first leg completed. As shown in the later sections, the research subjects of the five groups were physiological, biochemical or molecular-biological studies of hydrothermal vent- or cold seep-specific invertebrates (see Section 4), and thus we spent most time to collect live samples, e.g., deep-sea mussels, vesicomyid clams, polychaetes, and so on (see the pictures below). In addition, environmental parameters, including water temperature and sulfide concentration, of the mussel and polychaet colonies were analyzed. Some samples are also used for short-term rearing experiments. Other samples were kept alive and brought back to AORI, JAMSTEC, and Enoshima Aquarium for rearing experiments. Detailed analyses of genes, amino acids, and enzymes will be performed after the cruise. The preliminary reports of each group are in the section 4.



Mussels and polychaetes at Myojin Knoll (left) and young mussel colony at Suiyo Seamount (right)



Mussels (left) and vesicomyid clams at Off Hatsushima

2) Cruise Log (By Ms. Minamizawa)

	NT11-	09 Shipboard Log & Ship Track			
Date	Time	Description	Remarks	Position/Weather/ Wind/Sea condition (Noon)	
15Jun11	8:00	Scie} tists embark on Natsushima			
ISJUITT	9:00	Left YOKOSUKA for leaving point		12:00(GMT+9h)	
	10:30-11:00	Carried out onboard education & training for scientiscs		- 34-52.3N,139-39.6E Overcast	
	16:40-17:00	Konpira ceremony		 East-3(Gentle breeze) Sea smooth 	
	18:00-19:30	Scientific meeting			
16Jun11	3:30	Arrived at research area	Myojin knoll		
Iojunin	5:57	Released XBT	at 32-05.9100N, 139-51.5458E		
	7:23	Launched HPD on the surface			
	7:36	HPD dove & started het operation	HPD#1284	10-00(OMT+0+)	
	8:28	HPD landed on sea bottom (D=1342m)	32-06.288N, 139-51.996E	- 12:00(GMT+9h) 32-06.3N,139-52.1E	
	15:03	HPD left sea bottom (D=1243m)	32-06.206N, 139-52.048E	Overcast ESE-5 (Fresh breeze) See alight	
	15:40	HPD floated		 Sea slight 	
	15:57	Recovered HPD & finished her operation			
	16:10	Com'ced proceeding to Suiyo SMt.			
	19:00-19:30	Scientific meeting			
17Jun11	13:04	Released XBT	28-40.5694N, 140-36.4661E		
	15:00	Arrived at IZU OGASAWARA research area	Suiyo SMt.	- 12:00(GMT+9h) 28-47.2N,140-34.2E	
	19:00-19:30	Scientific meeting		Fine but cloudy SSW-4 (Moderate breeze) Sea smooth	
18Jun11	8:23	Launched HPD on the surface			
IoJuiii	7:15	HPD dove & started het operation	HPD#1285		
	9:25	HPD landed on sea bottom (D=1386m)	28-34.282N, 140-38.679E		
	11:28	HPD left sea bottom (D=1383m)	28-34.272N, 140-38.662E		
	12:07	HPD floated			
	12:24	Recovered HPD & finished her operation		12:00(GMT+9h) 28-34.3N, 140-38.7E	
	13:41	Launched HPD on the surface		Fine but cloudy	
	13:57	HPD dove & started het operation	HPD#1286	SSW-4 (Moderate breeze) Sea smooth	
	14:43	HPD landed on sea bottom (D=1386m)	28-34.285N, 140-38.668E		
	16:41	HPD left sea bottom (D=1383m)	28-34.281N, 140-38.682E		
	17:23	HPD floated			
	17:39	Recovered HPD & finished her operation			
	19:00-19:30	Scientific meeting			

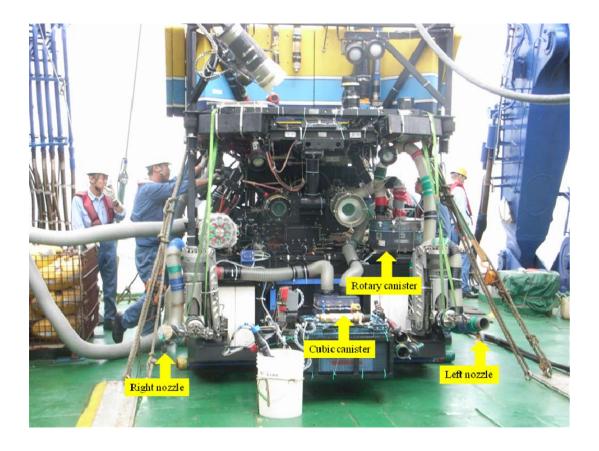
	8:30	Launched HPD on the surface		
19Jun11	8:43	HPD dove & started het operation	HPD#1287	
	9:32	HPD landed on sea bottom (D=1386m)	28-34.280N, 140-38.632E	
	14:25	HPD left sea bottom (D=1382m)	28-34.327N, 140-38.580E	 12:00(GMT+9h) 28-34.3N, 140-38.6E Fine but cloudy
	15:07	HPD floated		SSW-4 (Moderate breeze)
	15:24	Recovered HPD & finished her operation		Sea slight
	15:50	Com'ced proceeding to IZU OGASAWARA Myojin knoll		
	19:00-19:30	Scientific meeting		
20Jun11	15:30	Sighted Aoga-shima		
	19:00-19:30	Scientific meeting		12:00(GMT+9h) 31-55.3N,139-51.2E Overcast SSW-5 (Fresh breeze) Sea slight
	6:00	Arrived at dive point		
21Jun11	7:15	Launched HPD on the surface		
	7:26	HPD dove & started het operation	HPD#1288	
	8:14	HPD landed on sea bottom (D=1327m)	32-06.331N, 139-51.135E	
[10:21	HPD left sea bottom (D=1269m)	32-06.287N, 139-52.183E	
	11:00	HPD floated		
	11:15	Recovered HPD & finished her operation		12:00(GMT+9h) 32-06.3N,139-52.0E
	12:40	Launched HPD on the surface		Overcast
	12:50	HPD dove & started het operation	HPD#1289	SSW-5 (Fresh breeze) Sea slight
	13:38	HPD landed on sea bottom (D=1327m)	32-06.262N, 139-52.009E	
	15:12	HPD left sea bottom (D=1265m)	32-06.233N, 139-52.086E	
	15:48	HPD floated		
	16:02	Recovered HPD & finished her operation		
	16:20	Left research area for YOKOSUKA		
	18:00-18:30	Scientific meeting		
22Jun11	14:00	Arrived at YOKOSUKA		12:00(GMT+9h)
2200111	15:30	5 Scientists disembark from NATSUSHIMA		Fine but cloudy SSW-3 (Gentle breeze) Sea smooth
00 1	8:30	left YOKOSUKA for research area		
23Jun11	10:00-10:30	Carried out onboard education & training for scientiss		
	13:03	Released XBT	at 35-01.6N, 139-17.3E	
	14:20	Let go her anchor in 45m of water at Ito Ko		12:00(GMT+9h) 35-03.3N,139-27.5E
	15:15-15:45	Practiced boat, fire & collison drill at Ito Ko		Fine but cloudy SW-7 (Near gale) Sea moderate
	15:45-16:15	Practiced ISPS training for deter attacker brom approaching to ship at Ito Ko		
ļ	15:42	Left go her anchor with 3ss on deck		
	18:00-18:30	Scientific meeting		

24Jun11	6:15	Com'ced proceeding to research area		
	7:00	Arrived at dive point		
	8:26	Launched HPD on the surface		
	8:39	HPD dove & started het operation	HPD#1290	
	9:12	HPD landed on sea bottom (D=937m)	35-00.934N, 139-13.393E	
	11:03	HPD left sea bottom (D=855m)	35-00.956N, 139-13.329E	_ 12:00(GMT+9h)
	11:30	HPD floated		35-00.9N, 139-13.4E
	11:46	Recovered HPD & finished her operation		Fine but cloudy SW-6 (Strong breeze) Sea slight
	13:04	Launched HPD on the surface		Sea siight
	13:20	HPD dove & started het operation	HPD#1293	
	13:51	HPD landed on sea bottom (D=873m)	35-00.961N, 139-13.383E	
	16:01	HPD left sea bottom (D=854m)	35-00.955N, 139-13.322E	
	16:27	HPD floated		
	16:42	Recovered HPD & finished her operation		
25Jun11	8:32	Launched HPD on the surface		
20001111	8:47	HPD dove & started het operation	HPD#1292	
	9:26	HPD landed on sea bottom (D=937m)	35-00.054N, 139-13.545E	
	11:12	HPD left sea bottom (D=1176m)	35-00.072N, 139-13.470E	
	11:30	HPD floated		
	11:58	Recovered HPD & finished her operation		
	13:08	Launched HPD on the surface		
Γ	13:20	HPD dove & started het operation	HPD#1293	
	14:03	HPD landed on sea bottom (D=933m)	35-00.923N, 139-13.405E	
	16:01	HPD left sea bottom (D=854m)	35-00.959N, 139-13.332E	
	16:27	HPD floated		
	16:42	Recovered HPD & finished her operation		
26Jun11	9:30	Arrived at YOKOSUKA		
2030111		Scientists disembark from NATSUSHIMA		

3) Major equipments loaded to Hyper-Dolphin

a) Slurp Gun (Suction sampler)

It was used to collect benthos and fish. The nozzle attached to the left hand of the manipulator was connected to a rotary canister containing 6 bottles, which enable to keep samples from different points separated. Bottles were removed when necessary. In this cruise, another nozzle was set on the right hand, which was connected to another canister, which is cubic shape and contained no bottle.



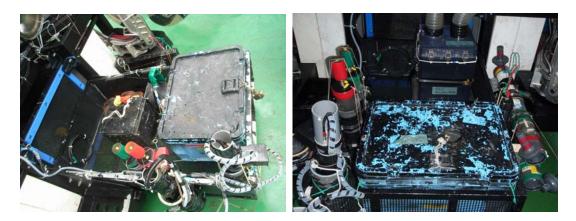
Arrangement of the slurp gun (suction sampler) in the front bay of Hyper-Dolphin



Cubic canister (left) and rotary canister (right)

b) Sample boxes

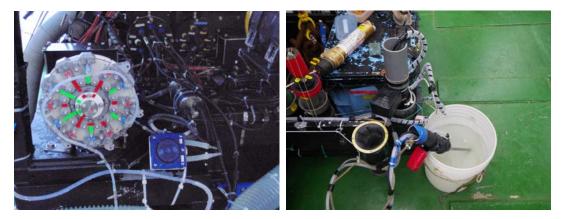
Two sample boxes were loaded in front of the vehicle. They were used to keep specimens collected by shovel and manipulator, and to bring traps



Sample boxes

c) Rotary clean seawater sampler (ROCS) and on-line thermometer

Rotary clean seawater sampler (ROCS) is a system to collect seawater. It consists of a nozzle, 6 polycarbonate bottles, and a pump. It is possible to collect seawater (up to 500 ml) at six specific sites by handling the nozzle with the manipulator. By setting an on-line thermometer in the nozzle, we measured the real-time temperature of the water.



Arrangement of ROCS

 Niskin water sampler, Van Dorn water sampler and MBARI corer Niskin samplers were used to obtain non-vent and non-seep seawater.
 Bandon sampler was used to collect vent and plume water.
 MBARI corers were used for sediment collection.



Niskin water sampler

MBARI corers



Van Dorn water sampler

e) Fish trap "Hodohodo-Kun"

Two types of trap were used to catch small fish (Hodohodo-Kun Nos. 1 & 2).



Two types of fish trap

4) Dive information (Dive point and track are in Appendix)

a) Dive 1284 (June 16, Myojin Knoll; Reporter, K. Inoue)

Equipments loaded: Suction sampler (Slurp gun) with a rotary canister containing 6-bottles, and that with a cubic canister, Rotary clean seawater sampler (ROCS), On-line sulfide sensor, On-line thermometer, Niskin water sampler, Sample Box, MBARI corer, Fish Trap "Hodohodo-Kun".

- 1. Water sampling using Niskin sampler at 600m depth
- 2. Water sampling using ROCS around an active chimney.
- 3. Sulfide monitoring around an active chimney.
- 4. Sediment sampling using MBARI corer
- 5. Sampling of mussels and polychaets using suction sampler
- 6. Sampling of chimney pieces
- 7. Measurement of the temperature of vent water at a chimney.
- 8. Taking picture of a mussel colony for long-range observation



Arrangement of research equipments at Dive #1284

b) Dive #1285 (June 18, 2011; Suiyo Seamount; Reporter, K. Inoue)

Equipments loaded: Suction sampler (Slurp gun) with a rotary canister containing 6-bottles, and that with a cubic canister, Rotary clean seawater sampler (ROCS), On-line sulfide sensor, On-line thermometer, Niskin water sampler, Sample Box, MBARI corer, Van Dorn water sampler, Fish Trap "Hodohodo-Kun" (No.1 & 2).

- 1. Water sampling using Niskin sampler at 600m depth
- 2. Sampling of vent water using Van Dorn sampler.
- 3. Fish traps were set.
- 4. Sampling of snails
- 5. Water sampling using ROCS around an active chimney.
- 6. Sulfide monitoring around an active chimney.
- 7. Mussel sampling.
- 8. Water sampling using Niskin sampler before leaving the bottom.

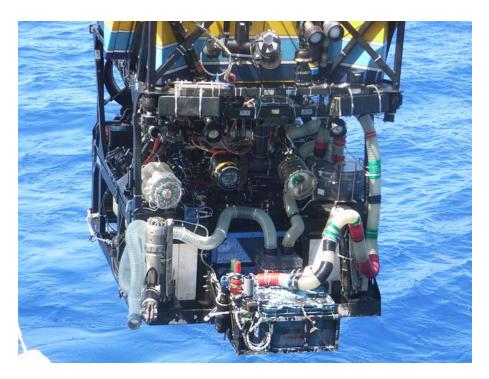


Arrangement of research equipments at Dive #1285

c) Dive #1286 (June 18, 2011; Suiyo Seamount; Reporter, K. Inoue)

Equipments loaded: Suction sampler (Slurp gun) with a rotary canister containing 6-bottles, and that with a cubic canister, Rotary clean seawater sampler (ROCS), On-line sulfide sensor, On-line thermometer, Niskin water sampler, Van Dorn water sampler, Sample Box, MBARI corer, Fish Trap "Hodohodo-Kun".

- 1. Mussel sampling
- 2. Recovery of traps.
- 3. Sampling of vent water using Van Dorn sampler.
- 4. Sampling of crabs.
- 5. Water temperature measurement around crabs
- 5. Water sampling using ROCS around an active chimney.
- 6. Sulfide monitoring around an active chimney and mussel colonies.

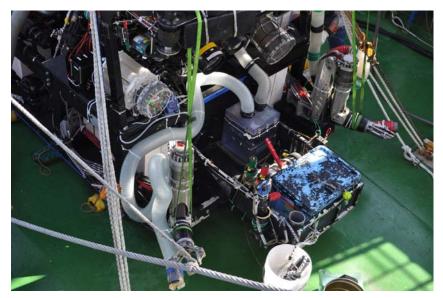


Arrangement of research equipments at Dive #1286

d) Dive 1287 (June 19, Suiyo Sea Mount; Reporter, T. Koito)

Equipments loaded: Suction sampler (Slurp gun) with a rotary canister containing 6-bottles, and that with a cubic canister, Rotary clean seawater sampler (ROCS), On-line sulfide sensor, On-line thermometer, Niskin water sampler, Van Dorn water sampler, Sample Box, MBARI corer, Fish Trap "Hodohodo-Kun".

- 1. Observation of some animals (octopus, Bathymodiolid mussels, etc..).
- 2. Sampling of Yunohana crub using suction sampler.
- 3. Water sampling using Van Dorn water sampler from a hydrothermal plume.
- 4. Sampling of crustacean using suction sampler.
- 6. Water sampling using ROCS around hydrothermal plume.
- 7. Sulfide monitoring around an active chimney.
- 8. Sediment sampling using MBARI corer
- 9. Sampling of mussels using suction sampler
- 10. Sampling of an octopus
- 11. Sampling of rock with bacterial mat
- 12. Observation of Central Cone of Suiyo Sea Mount



Arrangement of research equipments at Dive #1287

e) Dive 1288 (June 21, Myojin Knoll; Reporter, K. Inoue)

Equipments loaded: Suction sampler (Slurp gun) with a rotary canister containing 6-bottles, and that with a cubic canister, Rotary clean seawater sampler (ROCS), On-line sulfide sensor, On-line thermometer, Niskin water sampler, Sample Box, MBARI corer, Fish Trap "Hodohodo-Kun".

- 1. Failed to sample water, using Niskin sampler, at 600m because the shutter did not work.
- 2. Sampling of chimney pieces containing polychaetes.
- 3. Sampling of a chimney piece with barnacles
- 4. Sampling of polychaetes using suction sampler
- 5. Sampling of mussels using suction sampler
- 6. Water sampling using a Niskin sampler before leaving the bottom, but was not successful; the sample water did not remain.



Arrangement of research equipments at Dive #1288

f) Dive 1289 (June 21, Myojin Knoll; Reporter, K. Inoue)

Equipments loaded: Suction sampler (Slurp gun) with a rotary canister containing 6-bottles, and that with a cubic canister, Rotary clean seawater sampler (ROCS), On-line sulfide sensor, On-line thermometer, Niskin water sampler, Sample Box, MBARI corer, Fish Trap "Hodohodo-Kun".

- 1. Water sampling, using Niskin sampler, at 600m.
- 2. Cruise to the direction of point 6.
- 3. At a dead chimney, water sampling using ROCS.
- 4. Sulfide monitoring using the sulfide sensor.
- 5. Sampling of the chimney.
- 6. Cruise to find a small active chimney.
- 7. In front of the chimney, sediment sampling using a shovel.
- 8. Water sampling at a mussel colony using ROCS.
- 9. Sulfide sensing.
- 10. Water sampling using a Niskin sampler.
- 11. Sampling of mussels using suction sampler



Arrangement of research equipments at Dive #1289

g) Dive 1290 (June 24, Off Hatsushima; Reporter, Y. Nakamura)

Equipments loaded: Suction sampler (Slurp gun) with a rotary canister without 6-bottles, and that with a cubic canister, Rotary clean seawater sampler (ROCS), On-line sulfide sensor, On-line thermometer, Niskin water sampler, Sample Box (x1), MBARI corer, Fish Trap "Hodohodo-Kun", Shovel (Scoop sampler).

- 1. Water sampling using Niskin sampler at 500m depth.
- 2. Observation of a Calyptogena colony.
- 3. Water sampling using Niskin sampler at the bottom.
- 4. Sampling of Calyptogena with scoop sampler.
- 5. Observation of a Bathymodiolus colony.
- 6. Sampling of Bathymodiolus with suction sampler.
- 7. Sampling of Calyptogena with suction sampler.



Arrangement of research equipments at Dive #1290

h) Dive 1291 (June 24, Off Hatsushima; Reporter, Y. Nakamura)

Equipments loaded: Suction sampler (Slurp gun) with a cubic canister, Rotary clean seawater sampler (ROCS), On-line sulfide sensor, On-line thermometer, Niskin water sampler, Sample Box (x2), MBARI corer, Fish Trap "Hodohodo-Kun", Shovel (Scoop sampler).

- 1. Water sampling using Niskin sampler at 500m depth.
- 2. Observation of a mussel colony.
- 3. Water sampling using Niskin sampler at the bottme.
- 4. Water sampling using ROCS around a Bathymodiolus colony.
- 5. Measurement of the concentration of H2S in a Bathymodiolus colony.
- 6. Sampling of Bathymodiolus with suction sampler.
- 7. Water sampling using ROCS around a Calyptogena colony.
- 8. Measurement of the concentration of H2S in a *Calyptogena* colony.
- 9. Sampling of bottom mud with MBARI core x 2 (Green and Red).
- 10. Sampling of Fishes with fish trap "HodoHodo-kun".

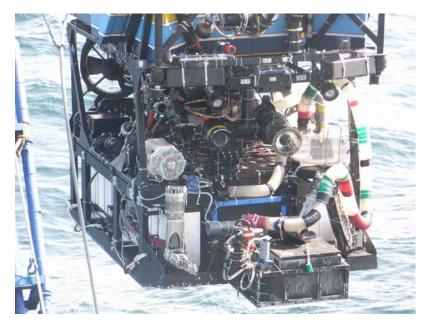


Arrangement of research equipments at Dive #1291

i) Dive 1292 (June 25, Off Hatsushima; Reporter, Y. Nakamura)

Equipments loaded: Suction sampler (Slurp gun) with a cubic canister, Rotary clean seawater sampler (ROCS), On-line sulfide sensor, On-line thermometer, Niskin water sampler, Sample Box (x2), MBARI corer, Fish Trap "Hodohodo-Kun", Shovel (Scoop sampler).

- 1. Water sampling using Niskin sampler at 500m depth.
- 2. Observation of a Calyptogena colony.
- 3. Water sampling using Niskin sampler at the bottqme.
- 4. Water sampling using ROCS around a Calyptogena colony.
- 5. Measurement of the concentration of H2S in a Calyptogena colony.
- 6. Sampling of Calyptogena with scoop sampler.
- 7. Sampling of Fishes with fish trap "HodoHodo-kun".



Arrangement of research equipments at Dive #1292

j) Dive 1293 (June 25, Off Hatsushima; Reporter, Y. Nakamura)

Equipments loaded: Suction sampler (Slurp gun) with a cubic canister, Rotary clean seawater sampler (ROCS), On-line sulfide sensor, On-line thermometer, Niskin water sampler, Sample Box (x2), MBARI corer, Fish Trap "Hodohodo-Kun", Shovel (Scoop sampler).

- 1. Observation of a Alaysia sp. colony.
- 2. Water sampling using ROCS around a Alaysia sp. colony.
- 3. Measurement of the concentration of H2S in a Alaysia sp. colony.
- 4. Sampling of Alaysia sp. with Hyper dolphin manipulator.
- 5. Sampling of Alvinocaris longirostris around a tubeworm colony with suction sampler.
- 6. Observation of a Lamellibrachia sp. colony.
- 7. Water sampling using ROCS around a Lamellibrachia sp. colony.
- 8. Measurement of the concentration of H2S in a Lamellibrachia sp. colony.
- 9. Sampling of Lamellibrachia sp. with Hyper dolphin manipulator.
- 10. Observation of a Calyptogena colony.
- 11. Measurement of the concentration of H2S in a Calyptogena colony.
- 12. Sampling of Calyptogena with scoop sampler.
- 13. Sampling of Fishes with fish trap "HodoHodo-kun".



Arrangement of research equipments at Dive #1293

4. RESEARCH INFORMATION

1) Studies on mechanisms of hypotaurine synthesis and its role in adaptation to toxic sulfide in hydrothermal vent-specific organisms

Koji Inoue¹, Tomoko Koito², Toshihiro Nagasaki¹, Azusa Kinjo¹, Yu Yamamoto¹, Wen Liu³, and Haruhiko Toyohara³

¹Atomosphere and Ocean Research Institute, The University of Tokyo, ²College of Bioresource and Sciences, Nihon University, ³Graduate School of Agriculture, Kyoto University

Background

The hydrothermal-vent specific animals must adapt to toxic hydrogen sulfide in vent water. A possible mechanism is the use of hypotaurine, which can react with sulfide and generate non-toxic thiotaurine. However, the pathway of hypotaurine synthesis has not been reported in marine invertebrates.

Methods and Results

In this cruise, we collected the deep-sea mussel *Bathymodiolus septemdierum* to study the hypotaurine synthesis pathway. During the cruise, we collected the mussels using suction sampler at Myojin Knoll and Suiyo Seamount. The mussels are dissected and frozen immediately after sampling or after exposure to certain conditions.

Future Plan

The mussels will be used for amino acid analyses and expression analyses of the enzyme genes involved in hypotaurine synthesis.

2) Culture experiment of Paralvinella hessleri in different tempgrature, and cDNA cloning of heat resistance relating gene.

Tsukasa Mori¹, Takao Yoshida², Shyuich Shigeno².

¹Laboratory of Marine Molecular Biochemistry, Department of Nihon University College of Bioresource Sciences.

²Japan Agency for Marine-Earth Science and Technology (JAMSTEC).

Background

Although annelid *Paralvinella grasslei* is known as a deep-sea vent endemic species that have heat tolerance, this tolerance might be greatly cooperation of cold water intake from gill. Therefore, we

exposed the relative spices of *Paralvinella grasslei* to the hot sea water in this experiment, and examined whether they can alive or not in the hot sea water.

Methods and Results

In this cruise, we collected the *Paralvinella hessleri* to study the metabolic analysis. During the cruise, we collected the whole body of the *Paralvinella hessleri* exposed hot sea water and cold sea water. These samples are frozen immediately after sampling, and stored in the -80C.

Future Plan

These samples will be used for cDNA synthesis and expression analyses for obtaining heat resistance relating gene.

3) Study on Symbiotic systems of deep-sea clam *Calyptogena* and intracellular sulfur oxidizing bacteria

Yoshimitsu Nakamura¹, Yuki Hongo¹, Akihiro Tame¹, Genki Ozawa¹, Daisuke Sekine¹, Ryusaku Deguchi² and Takao Yoshida¹ (JAMSTEC) ¹Japan Agency for Marine-Earth Science and Technology (JAMSTEC).

²Miyagi University of Education

Objective

Vesicomyid clams (*Calyptogena* spp.) are the dominant members in deep-sea chemosynthesis-based communities and harbor sulfur-oxydizing bacteria in the gill tissue cells. The non-functional degenerated digestive organs and the large size and structure of the gill of vesicomyids suggest that the hosts are not a filter feeder and rely on symbiot mediated autotrophic nutrition (Cavanaugh, 1983, Fiala-Medioni, 1984, 1986, Kennish, 1992). Genomic analyses of the symbionts as well as histological and ultrastructural observations of the host gill tissue endorse the hypothesis that endocellular chemoautotrophic bacteria play an important role in the nutrition of the clam (Fiala-Medioni, 1986, Kuwahara, 2007, 2008, Newton, 2007).

The symbionts of vesicomyids are housed only within the specific cell (bacteriocyte) in the gill tissue (Fiala-Medioni, 1986). It is assumed that the bacteriocytes peculiarly express the molecules associated with the symbiosis, so that the host raises the symbionts within the bacteriocytes. Though we recognize the importance of determining the bacteriocyte specific expressions, these examining approaches are still in their infancy. There are several reasons why it cannot be easily to identify the bacteriocyte specific expressions: uncultivable host and symbiont, many types of cells including bacteriocytes in the gill tissue

and a difficulty of the isolation of only bacteriocytes.

In this study we attempt to detect the molecules specifically expressed by the bacteriocyte in the gill tissue, utilizing histocytochemical approaches that is the direct detection assay system of bacteriocyte specific expressions by histocytochemical analysis using monoclonal antibody raised for the gill tissue.

Scientific Results

We collected *Calyptogena* sp. at the two sites of off Hatsushima Sagami Bay (Dive # 1290, 1291, 1293: 821m, 35°00.940' N, 139°13.247' E, and Dive # 1292: 1171m, 35°00.072' N, 139°13.503' E) and *Bathymodiolus septemdirum* (Dive # 1284: 1303m, 32°06.278' N, 139°52.081' E) by the ROV "Hyperdorphin" and dissected out the clams and mussels; gill, gonad, blood cells and other tissues. Then, the tissues to be cut out were immediately fixed in 4% paraformaldehyde or stored at -80°C.

Future works

In this cruse, we were successful in the sampling of fresh alive *Calyptogena* clams and *Bathymodiolus septemdirum*. We have plans for the investigations as follows:

- 1. Development of monoclonal antibody against bacteriosyte specific molecules.
- 2. in-situ analyses for the symbiotic specific gene expressions.
- 3. Tissue specific expression and localization of the symbiotic specific gene products.
- 4. Proteomics of the symbiotic bacteria.
- 5. Morphological and functional analysis of hemocytes.

4) Is it universal phenomenon for the hydrothermal vent community to detoxify hydrogen sulfide using thiotaurine? –Study on correlation between hydrogen sulfide concentration and the amount of amino acids.—

Tomoko Koito¹, Kei Sato², Shosei Yamagami³, Takuya Yahagi⁴ and Takefumi Yorisue⁴ ¹College of Bioresource Sciences, Nihon University, ²The University Museum, The University of Tokyo, ³Department of Earth Sciences, Okayama University, ⁴Atomosphere and Ocean Research Institute, The University of Tokyo

Background

It has been considered that the hydrothermal vent and cold seep endemic animals detoxify the ambient hydrogen sulfide to synthesize a non-toxic amino acid, thiotaurine using hypotaurine. However, it has not revealed that the correlation between concentration of hydrogen sulfide at the hydrothermal vent and the amount of thiotaurine in animals which inhabiting around the hydrothermal vent.

Methods and Results

In this cruise, we monitored the concentration of the hydrogen sulfide at Myojin Knoll and Suiyo Seamount which included active and non-active hydrothermal vents using hydrogen sulfide sensor and conducted water sampling at the same point using ROCS water sampler. Then, we collected chimney-attached animals at measuring hydrogen sulfide position, and counted the number of species, individuals and attached position of the chimney.

Future Plan

The collected animals will be used for amino acid and stable isotope analyses to reveal the correlation between the concentration of hydrogen sulfide and the amount of thiotaurine and the circulation of sulfur atom.

5) Study on immunity in the deep-sea hydrothermal vent mussels

Kazue Ohishi¹, Dai Sekine¹, Yoshimitsu Nakamura¹, Akihiro Tame², Yuki Hongo¹, Takao Yoshida¹, Sumihiro Koyama¹, and Tadashi Maruyama¹ ¹Japan Agency for Marine-Earth Science and Technology, ²Marine Works Ltd.Co.

Background

Inverterbrates possess innate immunity for protection against pathogens. The bivalve innate immune system is based on cellular immunity and soluble hemolymph factors. Blood cells are thought to play a central role in the innate immune system. The deep-sea mussels are found in association with large faunal communities living in chemosynthetic environments at the bottom of the sea floor, and they possess symbiotic bacteria in their gills.

Purpose and Objectives

Our final goal is to clarify what blood cells are involved in the innate immune system and how do the immune system distinguish the symbiont bacteria from pathogens. First, we aim to obtain the fundamental information of the immunity in the blood cells of the deep-sea mussles.

Materials and Methods

In this cruise, we sampled the deep-sea mussel *Bathymodiolus septemdierum*, *B. japonicus*, *and B. platifrons*. The blood cells were obtained by use a 5ml syringe and 23G needle. After dissection, tissue samples such as gills and mantle, were collected. The samples were immediately freezed or fixed, and

used for immunological analysis.

Research results

We observed some blood cells, which were morphologically and biochemically distinguishable. We are going to examine the function of these cells.

Future Plan

We will classify the blood cells in the deep-sea mussels, and examine the distribution in the whole mussel body and their role in the immunity.

5. ACKNOWLEDGMENTS

The science party expresses sincere thanks to the crew of RV Natsushima and the operation team of ROV Hyper-Dolphin.

6. NOTICE ON USING

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

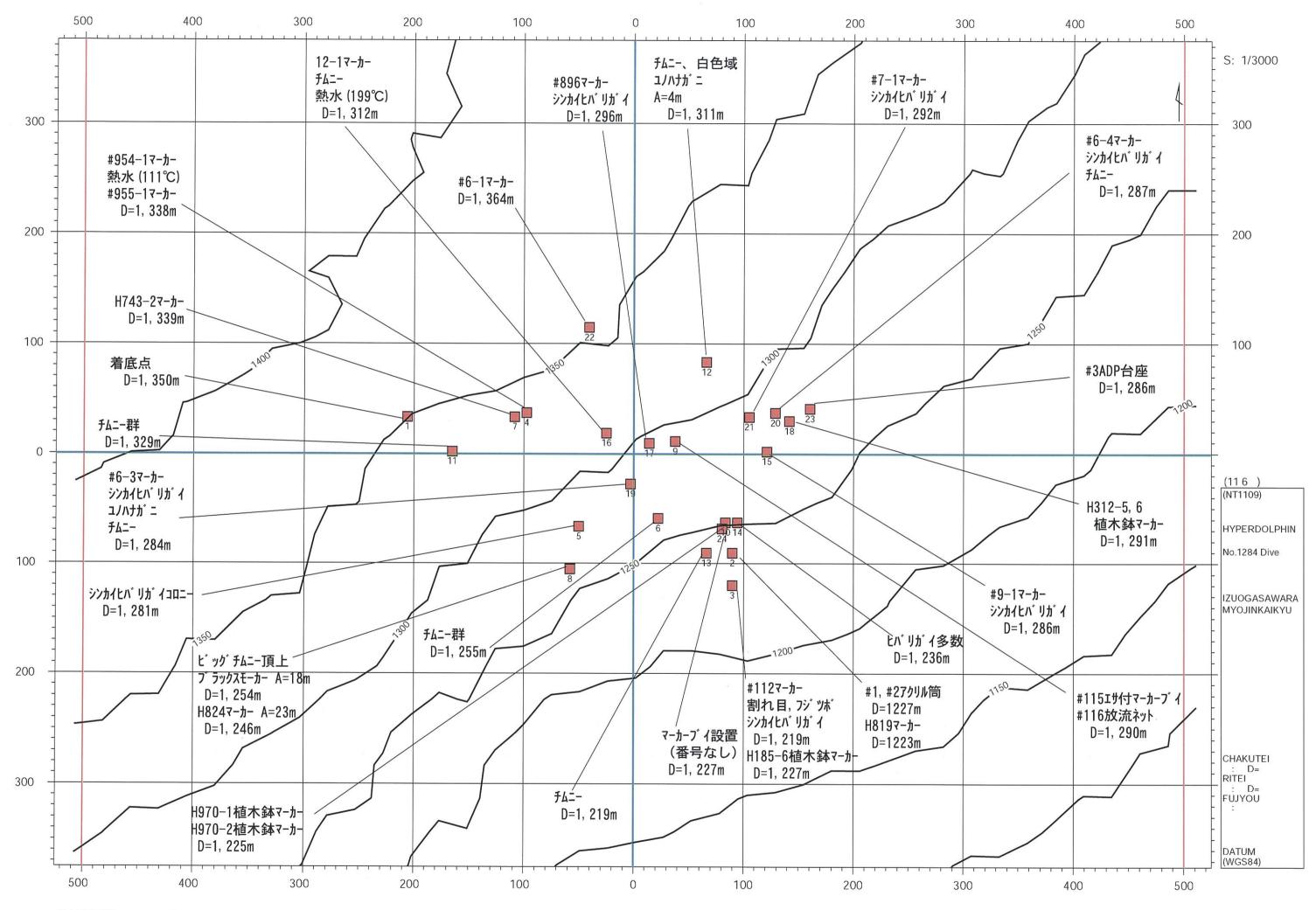
This cruise report is a preliminary documentation as of the end of the cruise.

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.

Appendix

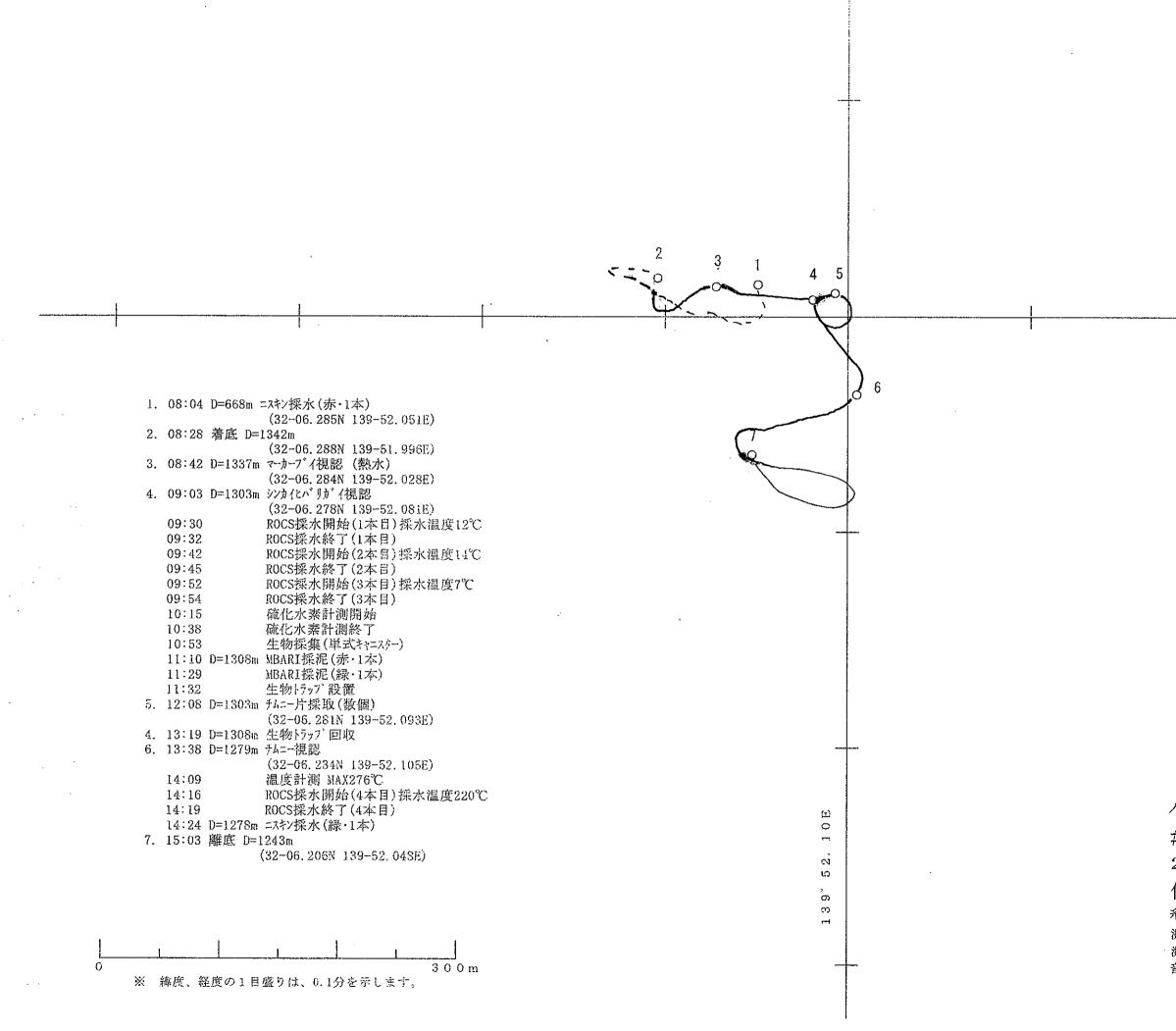
(Point maps and track charts)



XY ORIGIN 32-6.270N 139-52.100E

CENTER 32-6.270N 139-52.100E

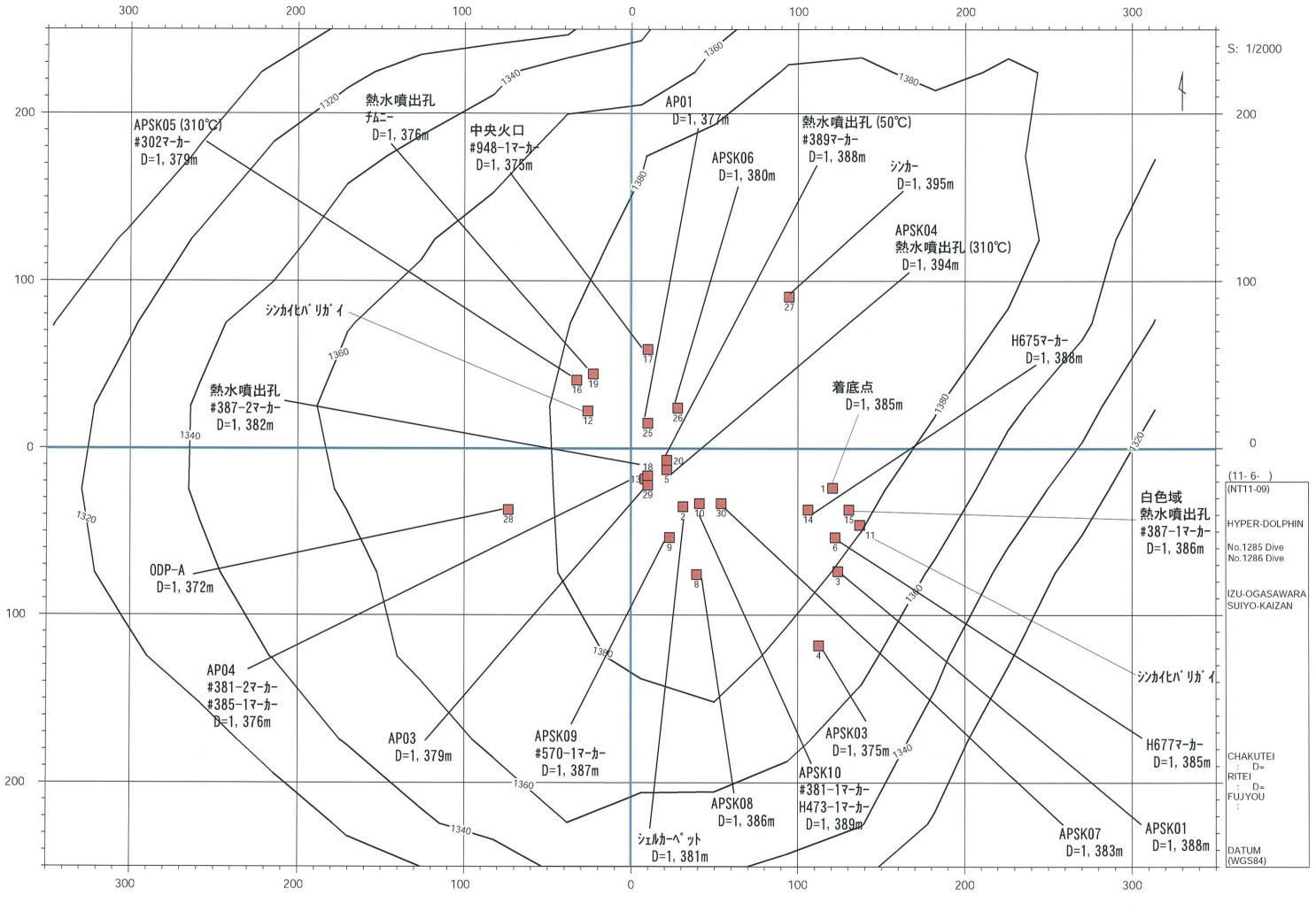
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32 06.27N

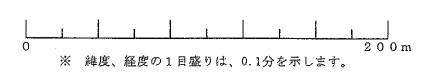
ハイパードルフィン3000 # 1 2 8 4 D I V E 2011年 6月16日 伊豆小笠原 明神海丘 統 尼 1 / 3000 測 位 D-GPS(Skyfix-XP Trimble SPS751) 測地系 WGS-84 DATUM (世界測地系) 音 速 1502.9m/s (D=1300m)

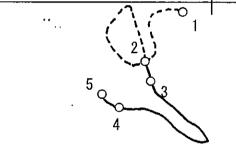


XY ORIGIN 28-34.300N 140-38.600E

CENTER 28-34.300N 140-38.600E

1.	09:00	D=637m =スキン採水(赤・1本)
		(28-34.297N 140-38.690E)
2.	09:25	着底 D=1386m
_	~~	(28-34. 282N 140-38. 677E)
3.	09:53	D=1382m バンドン採水(1本)
		(28-34. 276N 140-38. 679E)
	10:01	生物トラップ(ほどほどくん)1号,2号設置
	10:03	
4.	10:19	D=1381m 熱水噴出孔視認
		(28–34.268N 140–38.668E)
	10:21	H1285-1マーカー設置
	10:29	生物採集・岩石採取(#2キャニスター)
5.	10:46	温度計測(MAX105℃)
		(28-34.272N 140-38.662E)
	10:49	
	10:51	ROCS採水終了(1本目)
	10:59	ROCS採水開始(2本目)採水温度5℃
	11:03	
	11:12	
	11:24	
	11:28	離底 D=1383m



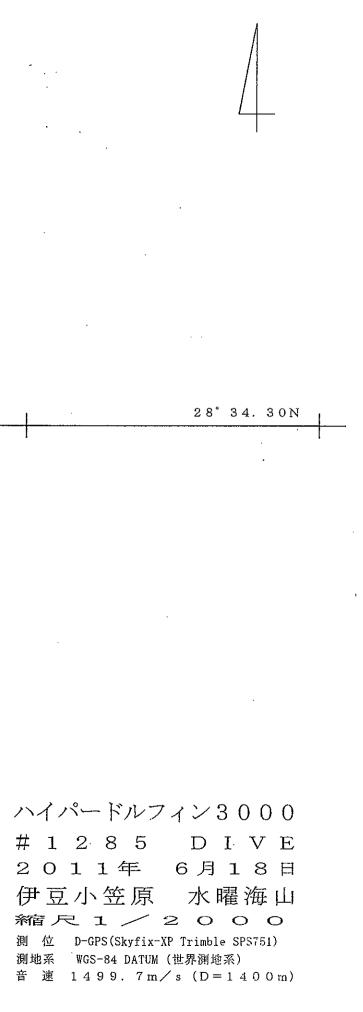


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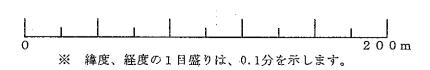
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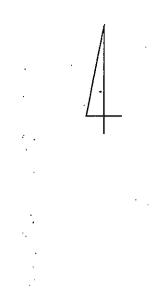


1.	14:43	着底 D=1386m
		(28–34.285N 140–38.668E)
2.	15:09	D=1384m 生物採集(多数・単式キャニスター)
		(28-34.275N 140-38.664E)
3.	15:17	D=1381m 生物トラップ(ほどほどくん)視認
		(28-34.281N 140-38.682E)
	15:35	生物採集(#2キャニスター)
		生物トラップ(ほどほどくん)1号,2号回収
	15:45	ベンドン採水(1本)
	15:58	生物採集(#3キャニスター)
	16:12	ROCS採水開始(3本目)採水温度26℃
	16:15	ROCS採水終了(3本目)
	16:23	ROCS採水開始(4本目)採水温度8℃
	16:25	ROCS採水終了(4本目)
	16:30	硫化水素計測開始
	16:36	硫化水素計測終了
	16:41	離底 D=1383m



140°38.60E

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.28°34.30N

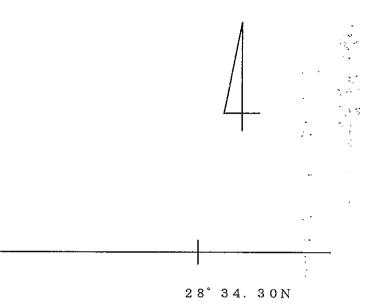
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ハイパードルフィン3000 #1286 DIVE 2011年 6月18日 伊豆小笠原 水曜海山 総 D-GPS(Skyfix-XP Trimble SPS751) 測地系 WGS-84 DATUM (世界測地系) 音速 1499.7m/ss(D=1400m)

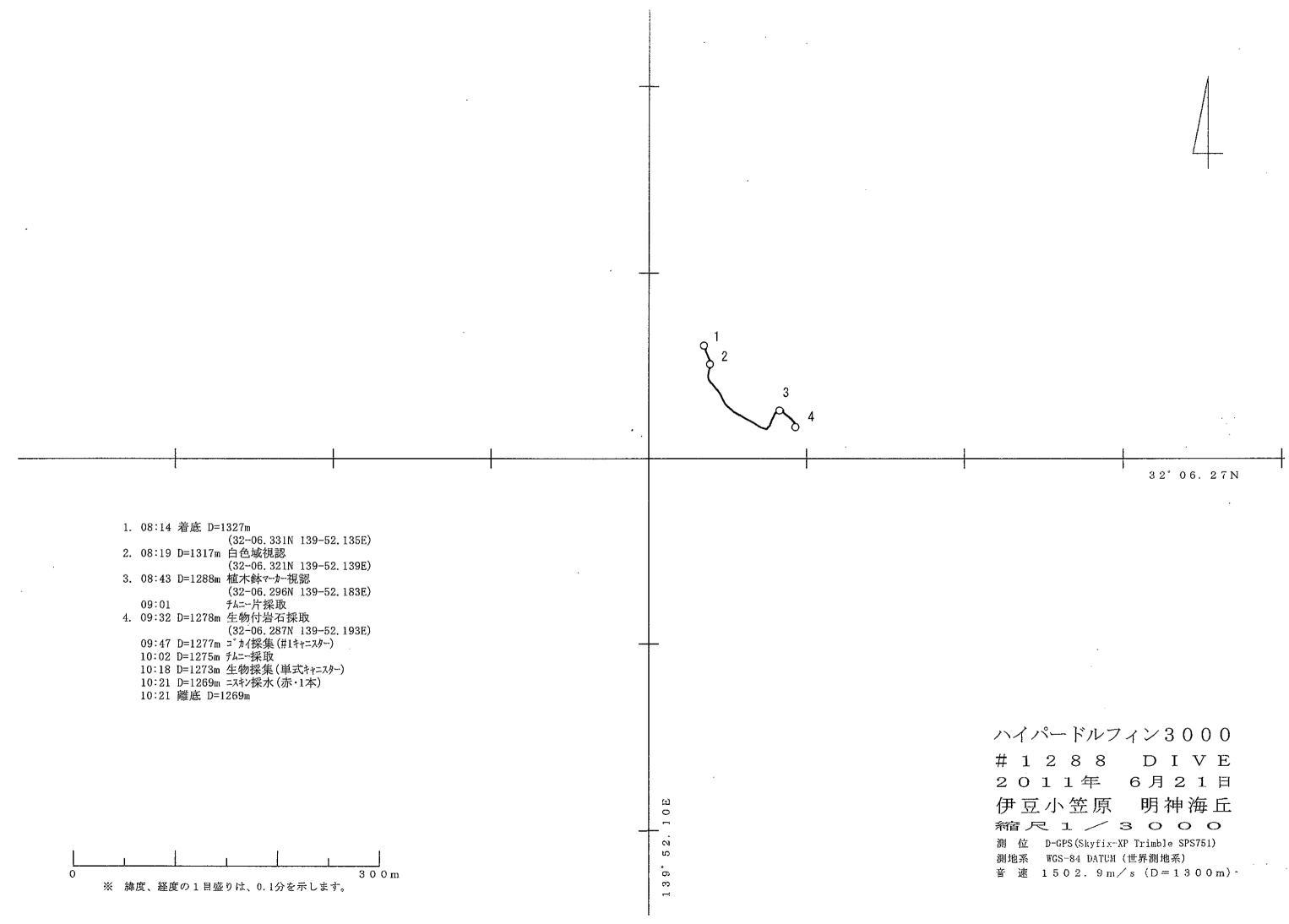
1. 09:32 着底 D=1386m	9 7 11
1. 09:02 / max, D 1000m (28-34.280N 140-38.632E) 2. 09:34 D=1384m ジュウモンジグゴ視認	$\gamma \gamma \gamma \gamma$
(28-34.273N 140-38.623E)	0 0 0 1 0 10
3. 09:42 D=1384m ジュウモンジダコ観察終了 (28-34.253N 140-38.608E)	5
09:56 生物採集(多数・#2キャニスター) 4. 10:11 D=1384m バンドン採水(1本)	
(28-34.263N 140-38.611E)	
5. 10:44 D=1385m #1287-1マーカー設置 (28-34.278N 140-38.624E)	
6. 10:49 D=1386m マーカー視認 (28-34.283N 140-38.627E)	4
7. 11:18 D=1387m 生物採集(#3キャニスター)	
(28-34.293N 140-38.611E) 11:30 ROCS採水開始(1本目)採水温度21℃	
11:33 ROCS採水終了(1本目) 11:43 ROCS採水開始(2本目)採水温度24℃	
11:46 ROCS採水終了(2本目)	
8. 12:12 D=1384m ROCS採水開始(3本目)採水温度24℃ (28-34.267N 140-38.609E)	
12:14 ROCS採水終了(3本目)	
12:20 ROCS採水開始(4本目)採水温度24℃ 12:22 ROCS採水終了(4本目)	
12:22 ROCS採水終了(4本目) 12:28 ROCS採水開始(5本目)採水温度22℃	
12:30 ROCS採水將了(5本目)	
12:40 ニスキン採水(赤・1本)	
12:43 ニスキン採水(緑・1本)	
12:54 MBARI採泥(禄·1本)	
13:02 生物採集(多数・単式キャニスター) 9. 13:13 D=1384m マーカー3個視認	
9. 13.13 D-1384m (28-34.281N 140-38.619E)	
10. 13:26 D=1388m MBARI採泥(赤・1本)	
(28-34. 282N 140-38. 641E)	
11. 13:32 D=1387m 夕コ視認 (28-34.292N 140-38.636E)	
13:36 /□採集(単式キャニスタ-)	
12. 13:50 D=1384m #307マーカー視認	
(28-34.317N 140-38.604E)	
13. 14:18 D=1382m 岩石1個採取 (28. 24. 207N 140. 29. 500F)	
(28-34.327N 140-38.580E) 14:25 離底 D=1382m	φ
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0 200m ※ 緯度、経度の1目盛りは、0.1分を示します。	4
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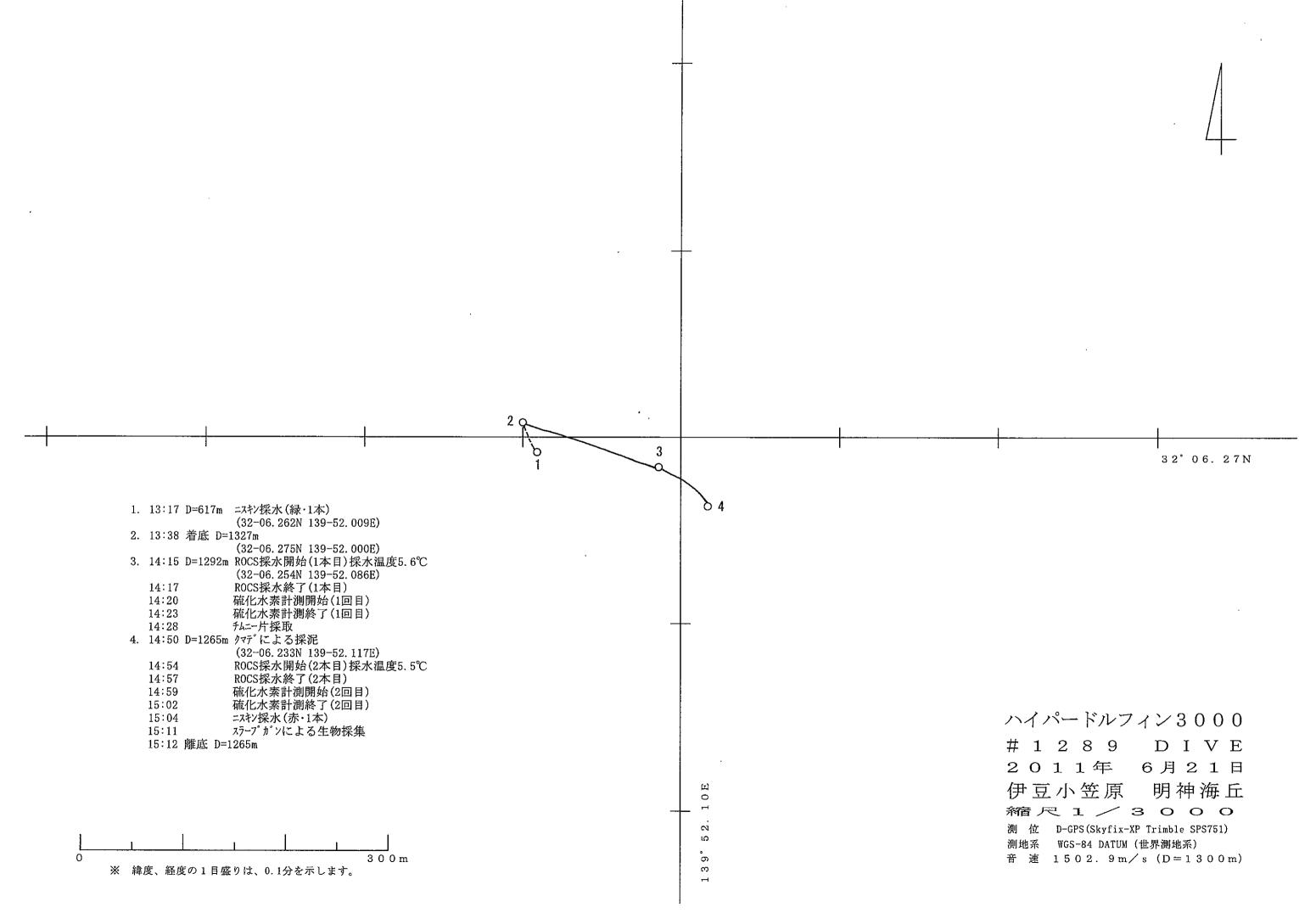
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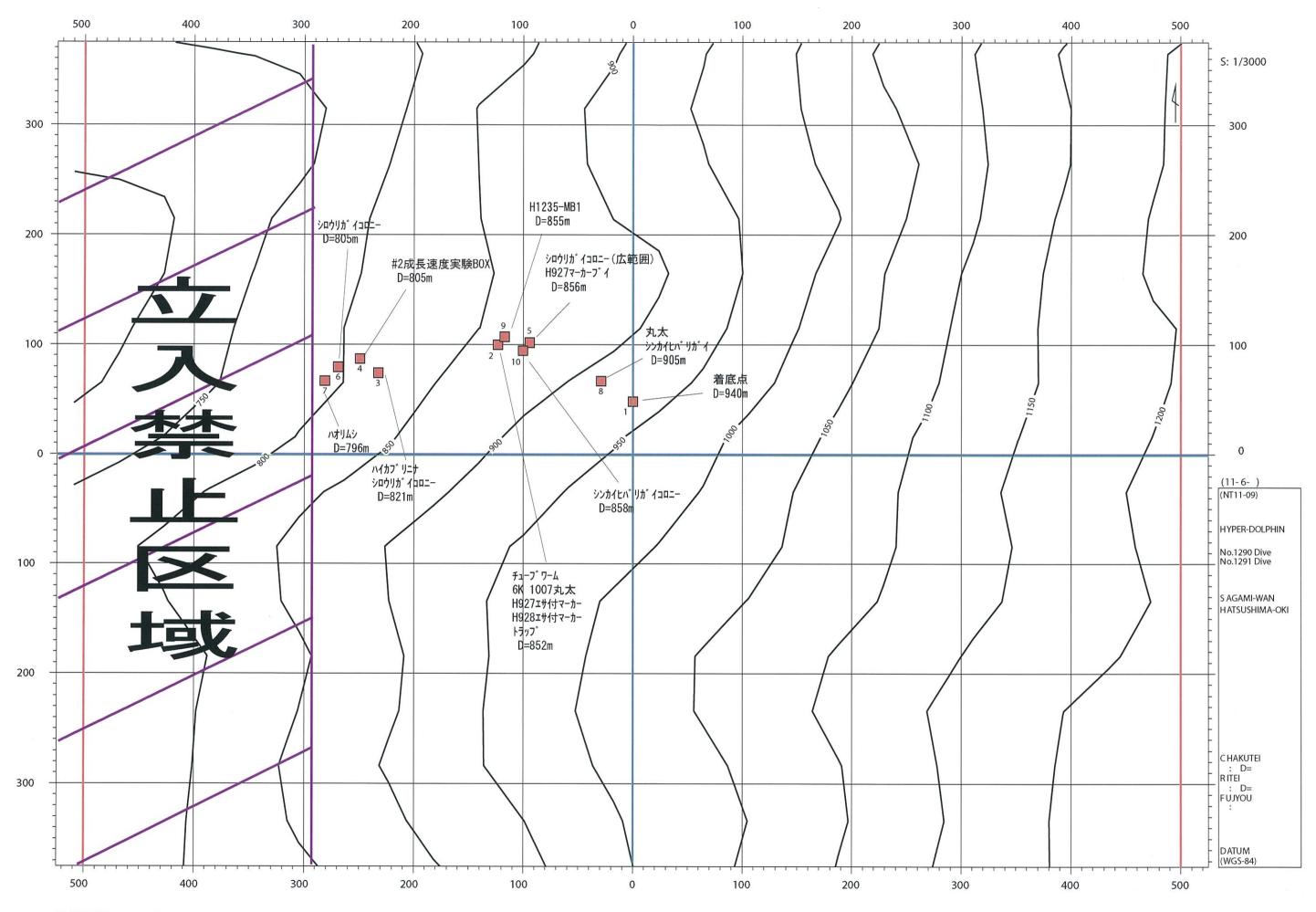


ハイパードルフィン3000 #1287 DIVE 2011年 6月19日 伊豆小笠原 水曜海山 統配尺1/2000 測位 D-GPS(Skyfix-XP Trimble SPS751) 測地系 WGS-84 DATUM (世界測地系) 音速 1499.7m/s (D=1400m) 1 e

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XY ORIGIN 35-0.900N 139-13.400E

CENTER 35-0.900N 139-13.400E

9 D=510m ニスキン採水(赤・1本)	
(35-00.934N 139-13	3, 393E)
2 着底 D=937m	
(35-00,931N 139-13	3.412E)
6 D=921m ニスキン採水(緑・1本)	
(35-00.934N 139-1)	3. 388E)
5 シロウリガイ採集	
6 H1290マーカー設置	
1 D=913m 植木鉢マーカー視認	
	〔式キャニスター(大)・多数)
///	
	3. 329E)
2 チューフ゛ワーム採集	
3 離底 D=855m	
5 ジャウリカ [*] イ採集 6 H1290マーカー設置 1 D=913m 植木鉢マーカー視認 (35-00.937N 139-1: 8 ジンカイヒハ [*] リカ [*] イ採集(単式キャ (35-00.956N 139-1: 2 チューフ [*] ワーム採集	3.382E) 式キャ=スター(大)・多数 /=スター(小))

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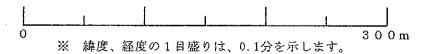
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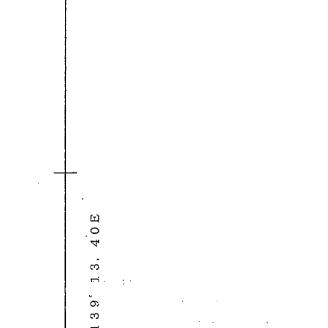
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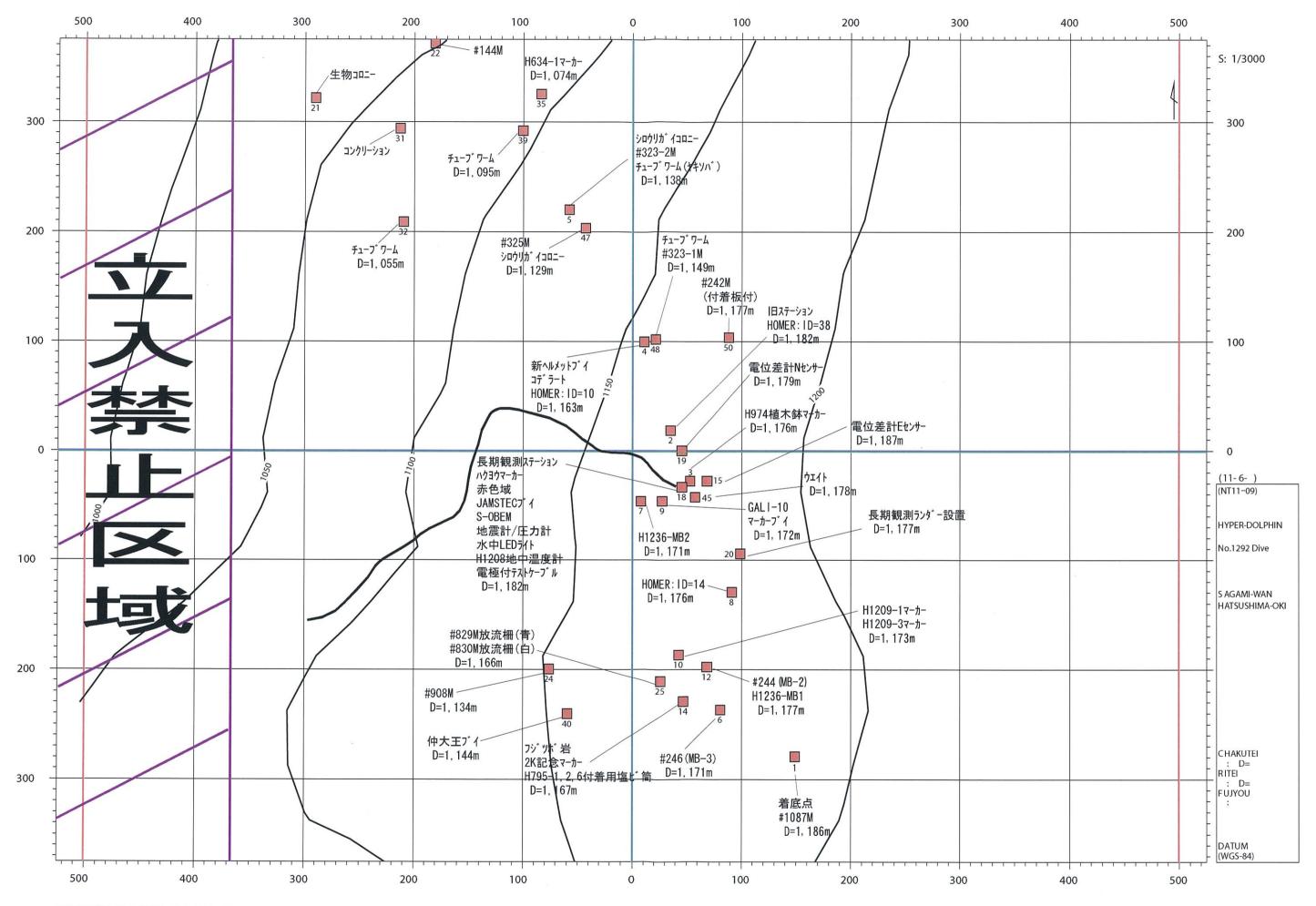
ハイパードルフィン3000 # 1 2 9 0 D I V E 2 0 1 1 年 6 月 2 4 日 木目 枝 湾 初 島 沖 絲病 尺 1 ~ 3 0 0 0 測 位 D-GPS(Skyfix-XP Trimble SPS751) 測地系 WGS-84 DATUM (世界測地系) 音 速 1 4 8 8.4 m/s (D=1000m)

 1. 13:40 D=511n =スキン探水(赤・1本) (35-00.961N 139-13.383E) 2. 13:51 着底 D=873m (35-00.946N 139-13.355E) 3. 14:02 D=857m 高度をとって移動 (35-00.947N 139-13.326E) 4. 14:17 D=910m =スキン採水(緑・1本) (35-00.940N 139-13.380E) 14:23 ROCS採水開始(1本目)採水温度4. 14:25 ROCS採水熱了(1本目) 14:27 硫化水素計測開始(1回目) 14:30 硫化水素計測開始(1回目) 14:39 ジカイレ[*]リガ[*] イ採集(単式キャニスター) 14:48 D=909m ROCS採水開始(2本目)採水温度4. 14:55 ROCS採水熱了(2本目) 14:57 ROCS採水熱了(3本目) 14:59 硫化水素計測開始(2回目) 15:66 硫化水素計測開始(2回目) 15:31 D=855m ROCS採水開始(4本目)採水温度4. (35-00.955N 139-13.322E) 15:33 ROCS採水熱了(4本目) 15:36 硫化水素計測器力(3回目) 15:39 硫化水素計測器力(3回目) 15:44 MBARI採泥(緑・1本) 15:48 生物トラッフ*(ほどほどくん)設置 15:56 /ァマテ*による採泥 16:01 離底 D=854m 	4℃ 5℃		
	 	139°13.40E	

ハイパードルフィン3000 #1291 DIVE 2011年 6月24日 相模湾 初島 沖 統存 尺 1~3000 測 位 D-GPS(Skyfix-XP Trimble SPS751) 測地系 WGS-84 DATUM (世界測地系) 音速 1488.4m/s (D=1000m)

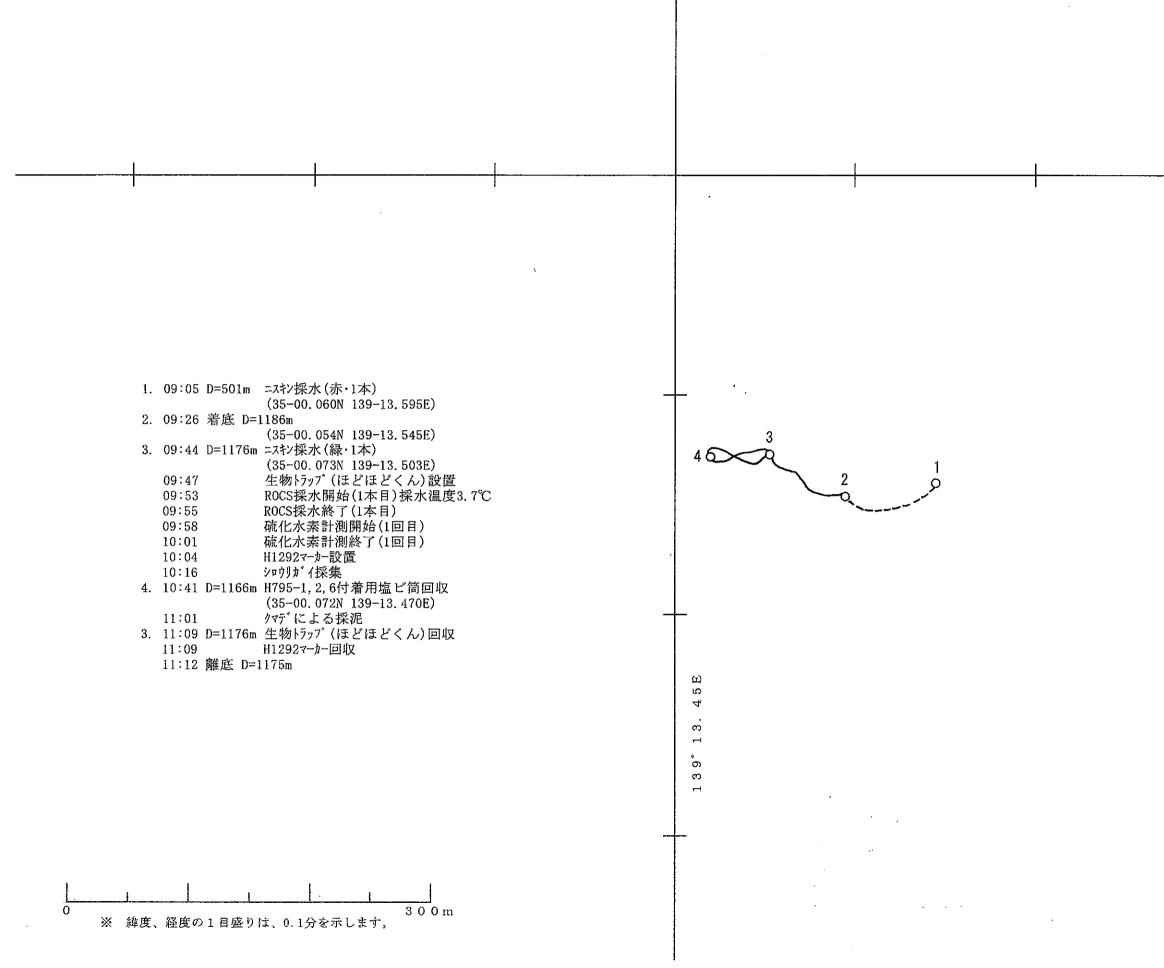
35°00.90N





XY ORIGIN 35-0.200N 139-13.450E







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35°00.20N

ハイパードルフィン3000 # 1 2 9 2 D I V E 2 0 1 1 年 6 月 2 5 日 木目 枝 湾 初島 沖 新宿 尺 1 3 0 0 0 測 位 D-GPS(Skyfix-XP Trimble SPS751) 測地系 WGS-84 DATUM (世界測地系) 音 速 1486.6m/s (D=1300m)

	5 $0 $ $4 $ $3 $ $2 $ $0 $ 1	
 14:03 着底 D=933m (35-00.923N 139-13.405E) 14:18 D=928m ROCS探水開始(2本目)採水温度4.3°C (35-00.926N 139-13.396E) ROCS探水終了(2本目) 14:22 硫化水素計測開始(1回目) 14:26 硫化水素計測線了(1回目) 14:31 スラーブがンによる生物採集 14:53 D=899m ROCS採水開始(3本目)採水温度4.5°C (35-00.936N 139-13.377E) 14:55 D=8899m ROCS採水開始(3本目) 14:57 硫化水素計測開始(2回目) 15:00 硫化水素計測開始(3回目) 15:06 チューブアー払採集 15:07 硫化水素計測開始(3回目) 15:10 硫化水素計測解分(3回目) 15:25 D=856m 生物トラッブ(ほどほどくん)設置 (35-00.954N 139-13.337E) 15:34 ケマデによる採泥 (35-00.959N 139-13.332E) 15:57 D=855m ケマデ(による探泥 (35-00.959N 139-13.332E) 16:03 離底 D=855m 	1 139, 13, 40 1 1 1 3 9, 1 3 9, 1 9 1 3 9, 1 9 1 3 9, 1 9 1 3 9, 1 9 1 9, 1 9 1 9, 1 9, 1 9, 1 9, 1 9	、

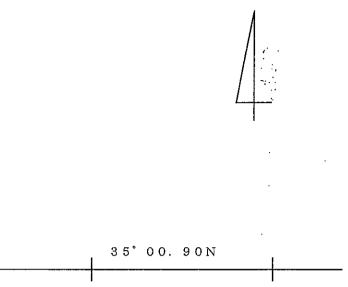
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ハイパードルフィン3000 # 1 2 9 3 D I V E 2 0 1 1 年 6 月 2 5 日 相 枝 湾 初 島 沖 縮 尺 1 3 0 0 0 側 位 D-GPS(Skyfix-XP Trimble SPS751) 側地系 WGS-84 DATUM (世界測地系) 音 速 1486.6m/s (D=1300m)