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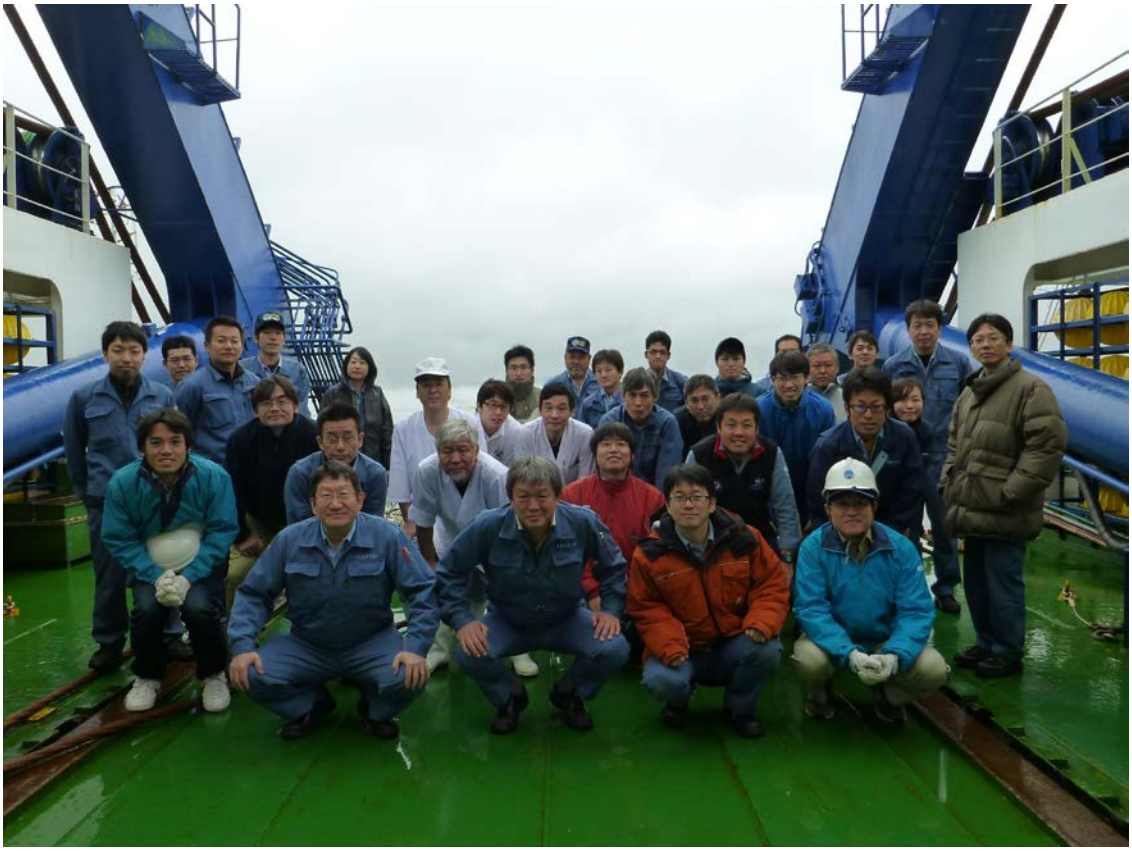
YK16-16

(10– 25 November 2016)

Preliminary Cruise Report

Response of marine ecosystem to the ocean acidification in the subarctic western North Pacific & Tsunami early warnings by the seafloor electromagnetic observations



The subarctic western North Pacific and North Pacific Basin

YOKOSUKA

December 2016

JAMSTEC

Note

This cruise report is a preliminary documentation published in approximately a month after the end of this cruise. It may not be corrected even if changes on contents 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 principal investigator and parsons in charge of respective observations for the latest information and permission before using. Users of data are requested to submit their results to JAMSTEC Data Management Group (DMG).

Acknowledgments

We are grateful to the captain and crew of the R/V ‘‘Yokosuka’’ for their support during the cruise.

December 2016

Chief Scientist of YK16-16

Tetsuichi Fujiki
JAMSTEC

Cruise Report ERRATA of the Nutrients part

page	Error	Correction
28	potassium nitrate CAS No. 7757-91-1	potassium nitrate CAS No. 7757-79-1
26	1N H_2SO_4	1M H_2SO_4

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1. Outline of YK16-16

1.1 Cruise information

Cruise ID: YK16-16

Research vessel: YOKOSUKA

Cruise title: Response of marine ecosystem to the ocean acidification in the subarctic western North Pacific & Tsunami early warnings by the seafloor electromagnetic observations

Cruise period (port call): 10 November (Harumi, Tokyo) – 25 November 2016 (Yokosuka, Kanagawa)

Research area: The subarctic western North Pacific and North Pacific Basin

Chief Scientist: Tetsuichi Fujiki (Research and Development Center for Global Change (RCGC), JAMSTEC)

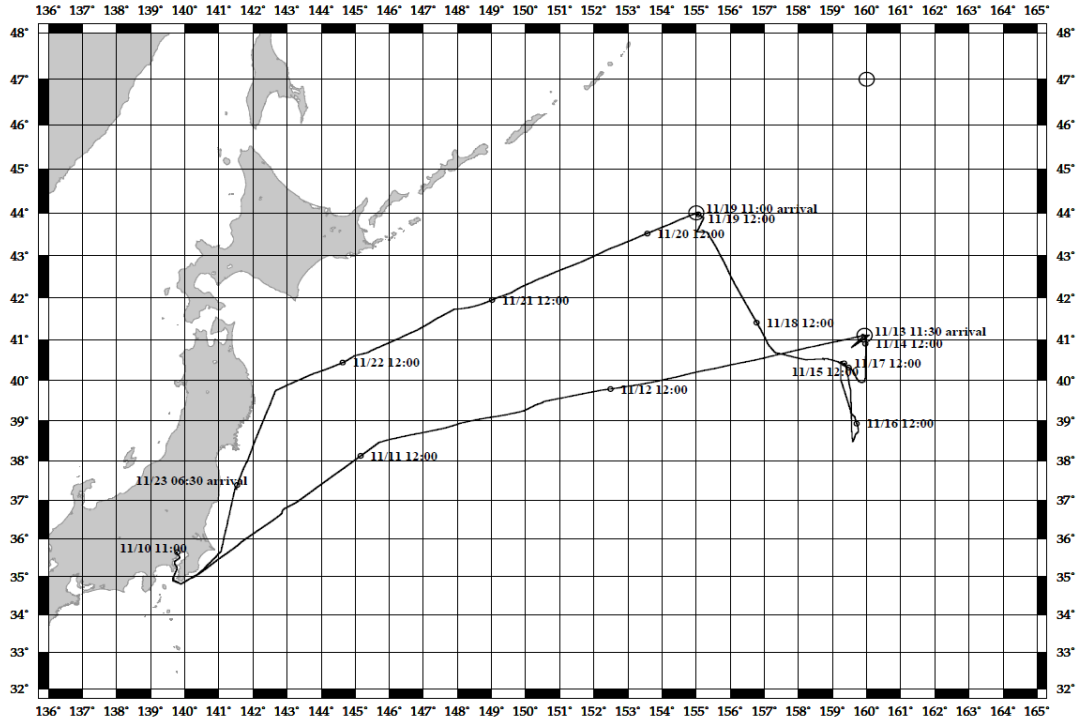
Deputy Chief Scientist: Hiroaki Toh (Graduate School of Science, Kyoto University)

Representative of the Science Party: Tetsuichi Fujiki (RCGC, JAMSTEC)
Hiroaki Toh (Kyoto University)

Proposal title:

1. Response of marine ecosystem to the ocean acidification in the subarctic western North Pacific
2. Tsunami early warnings by the seafloor electromagnetic observations

Cruise truck:



* We couldn't go to the time-series station K2 (47°N, 160°E) during the cruise period due to bad weather.

Cruise log:

Date	Local Time	Note	Position/Weather/Wind/Sea condition
10-Nov-16	09:00	Scientists onboard.	Tokyo Bay
	10:00	Let go all shore lines & left HARUMI for research area K2(North west pacific).	35-32.6N, 139-50.6E
	13:00~14:00	Carried out Shipboard education for scientist.	Cloudy
	14:00~15:00	Scientist meeting.	NE-4 (Moderate breeze)
			2 (Sea Smooth)
			1 (Low Swell Short)
			Visibly: 8'
11-Nov-16	9:00~10:00	Carried out traning for scientist.	East-off KINKA-SAN
	13:00~14:00	Scientist meeting.	38-07.1N, 145-09.4E
	14:00	Changed the destination to St.NWP	Overcast
			SE-4 (Moderate breeze)
			3 (Sea Slight)
			1 (Low Swell Short)
			Visibly: 8'
12-Nov-16	00:00	Put ship's clock ahead 60' min for S.M.T. in long.	Western North Pacific
			39-47.5N, 152-29.5E
			Cloudy
			ESE-5 (Fresh breeze)
			4 (Sea Moderate)
			1 (Low Swell Short)
			Visibly: 8'
13-Nov-16	11:30	Arrived at research area (North west pacific) .	Western North Pacific
	12:33	launched transducer Cheak signal SFEMS.	41-07.2N, 159-53.9E
	12:55	Hoisted up transducer.	Overcast
	13:17-14:55	Carried out Norpac Net operation 6cast.	SE-5 (Fresh breeze)
	15:07-15:30	Carried out FRRF operation 1cast.	4 (Sea Moderate)
			2 (Low Swell Slong)
			Visibly: 8'
14-Nov-16	03:37	Sent out release command to SFEMS.	Western North Pacific
	07:33	Refloted SFEMS.	40-53.9N, 159-58.1E
	08:00	Recovered SFEMS.	Cloudy
	10:12-11:06	Carried out CTD operation.	WSW-6 (Strong breeze)
	11:30	Let's research area (st.NWP) for south ward.	5 (Sea Rough)
			4(Moderate Average)
			Visibly: 8'
15-Nov-16	17:45	Finished heave to & proceeded to south ward.	Western North Pacific
			40-19.0N, 159-28.7E
			Fine but Cloudy
			South-3 (Gentle breeze)
			3 (Sea Slight)
			4(Moderate Average)
			Visibly: 8'
16-Nov-16	00:15	Com'ced heave to.	Western North Pacific
			38-55.7N, 159-42.7E
			Rain
			SW-6 (Strong breeze)
			6(Very rough)
			7(Heavy Swell Average)
			Visibly: 2'
17-Nov-16	All day	Ward off heavy weather.	Western North Pacific
			40-25.0N, 159-20.2E
			Rain
			West-8(Gale)
			7(High)
			7(Heavy Swell Average)
			Visibly: 8'

Date	Local Time	Note	Position/Weather/Wind/Sea condition
18-Nov-16	07:00	Finished heave to & proceeded to research area (St.KNOT) .	Western North Pacific
			41-24.5N, 156-46.6E
			Fine but Cloudy
			NW-6 (Strong breeze)
			5 (Sea Rough)
			4(Moderate Average)
			Visibly: 8'
19-Nov-16	02:30	Com'ced heave to.	Western North Pacific
	08:30	Finished heave to & proceeded to research area (St.KNOT) .	43-57.5N, 155-03.8E
	11:00	Arrived at reserch area(St.KNOT)	Rain
	14:10-16:20	Carried out CTD operation.	WSW-6 (Strong breeze)
	17:51-18:35	Carried out CTD operation.	5 (Sea Rough)
	20:15-21:18	Carried out VMPS operation.(2 cast)	4(Moderate Average)
	22:07-23:51	Carried out Norpac Net operation.(10 cast)	Visibly: 8'
20-Nov-16	00:03-00:23	Carried out FRRF operation.	Western North Pacific
	01:21-05:18	Carried out CTD operation.(4 cast)	43-31.0N, 153-34.1E
	05:30	Left reserch area for yokosuka.	Rain
	11:38-11:58	Carried out figure eight turn measurement.	South-5 (Fresh breeze)
			3 (Sea Slight)
			2 (Low Swell Slong)
			Visibly: 8'
21-Nov-16		Com'ced proceeding to YOKOSUKA.	Western North Pacific
			41-57.1N, 149-01.0E
			Fine but Cloudy
			WNW-7 (Near gale)
			6 (Very rough)
			7 (Heavy Swell Average)
			Visibly: 8'
22-Nov-16		Com'ced proceeding to YOKOSUKA.	Off sanriku
			40-26.2N, 144-37.9E
			Rain
			NNW-5 (Fresh breeze)
			4 (Sea Moderate)
			1 (Low Swell Short)
			Visibly: 4'
23-Nov-16	06:30	Arrived at off FUKUSHIMA.	Off fukushima
	06:33	Released XBT at (37-22.4226N 141-32.2788E)	37-19.8N, 141-30.1E
	06:55	Com'ced MBES mapping survey.	Fine but Cloudy
	16:13	Released XBT at (37-17.2169N 141-28.4468E)	North-5 (Fresh breeze)
	17:06	Finished MBES mapping survey.	4 (Sea Moderate)
	17:10	Left off FUKUSHIMA for YOKOSUKA.	3 (Moderate Short)
			Visibly: 8'
24-Nov-16	10:45	Stationed for anchoring.	YOKOSUKA sec. 4
	11:00	Let go her port anchor in 31.0m of water, arrived at YOKOSUKA sec. 4.	35-19.7N, 139-40.9E
			Fog
			NNW-6 (Strong breeze)
			3 (Sea Slight)
			1 (Low Swell Short)
			Visibly: 1'
25-Nov-16	09:00	Arrived at YOKOSUKA(JAMSTEC), then completed voy. No.YK16-16.	

1.2 Cruise participants

Scientists

TETSUICHI FUJIKI	Japan Agency for Marine-Earth Science and Technology
MINORU KITAMURA	Japan Agency for Marine-Earth Science and Technology
KATSUNORI KIMOTO	Japan Agency for Marine-Earth Science and Technology
MASAHIDE WAKITA	Japan Agency for Marine-Earth Science and Technology
CHISATO YOSHIKAWA	Japan Agency for Marine-Earth Science and Technology
KEISUKE SHIMIZU	Japan Agency for Marine-Earth Science and Technology
HIROAKI TOH	Graduate School of Science, Kyoto University
YOSHIHISA MINO	Institute for Space-Earth Environmental Research, Nagoya University
MAKOTO SUGIMURA	Enoshima Aquarium
TAKUTO MINAMI	Earthquake Research Institute, University of Tokyo

Research Engineers

SHUNGO OSHITANI	Marine Works Japan, Ltd.
SHINSUKE TOYODA	Marine Works Japan, Ltd.
KEITARO MATSUMOTO	Marine Works Japan, Ltd.
TOMOMI SONE	Marine Works Japan, Ltd.
HIROYUKI HAYASHI	Nippon Marine Enterprises, Ltd.

R/V YOKOSUKA Crews

Captain	SHINYA RYONO
Chief Officer	AKIHISA TSUJI
1st Officer	TAKESHI MURAMATSU
2nd Officer	SHOZO FUJII
3rd Officer	YUKI ITO
Chief Engineer	MINORU TSUKADA
1st Engineer	SHINITI IKUTA
2nd Engineer	KATSUTO YAMAGUCHI
3rd Engineer	KAZUKI ONO
Chief Electronics Operator	TAKEHITO HATTORI
2nd Electronics Operator	TOSHIHIKO YUASA
Boat Swain	HATSUO ODA
Quarter Master	KANAME HIROSAKI
Quarter Master	YUKITO ISHII
Quarter Master	KOSEI KAWAMURA
Quarter Master	SHINYA UENO

Sailor	YUTA OHJIRI
Sailor	TOMOKI ASAKUNI
No.1 Oiler	JUNJI MORI
Oiler	KATSUYUKI MIYAZAKI
Oiler	RYOTA SUZUKI
Oiler	SHOTA SHIMOHATA
Assistant Oiler	SEIYA WATANABE
Chief Steward	SUETO SASAKI
Steward	HIRONOBU HODOKUMA
Steward	TSUYOSHI NAGATOMO
Steward	MASANAO KUNITA
Steward	SEIYA MATSUMOTO

1.3 Research brief

1.3.1 Response of marine ecosystem to the ocean acidification in the subarctic western North Pacific

The subarctic western North Pacific is a cyclonic upwelling gyre (western subarctic gyre; WSG) that extends from the northeast of Japan to near the international dateline. To investigate the spatial and temporal variability of biogeochemical processes in the WSG, time-series observations have been carried out since 1997 at time-series stations KNOT (44°N, 155°E) and /or K2 (47°N, 160°E), indicating that ocean acidification was rapidly progressing in this gyre. However, the effect of ocean acidification on lower trophic levels in this region is not well understood.

The main purpose of this research is to investigate the plankton community response to the progress of ocean acidification in the WSG. During the cruise period, we couldn't go to Sta. K2 due to bad weather. However, we conducted the following main studies at Sta. KNOT.

Content of research:

(a) Impact assessment of ocean acidification on marine organisms based on dissolved chemical constituents
(b) Relationship between plankton community and ocean acidification
(c) Measurements of carbonate shell density of planktic foraminifers and pteropods by the Micro-focus X-ray CT
(d) Increase of N_2O production rates as a consequence of ocean acidification

Observations and operations:

(1) CTD cast and water sampling/biochemical analysis
(2) Plankton sampling by using the VMPS and NORPAC nets
(3) Assessment of phytoplankton photosynthesis by fast repetition rate fluorometry

(4) On-deck ^{13}C incubation experiments

(5) Upper ocean current measurements by shipboard ADCP

(6) Sea surface water sampling

1.3.2 Tsunami early warnings by the seafloor electromagnetic observations

We succeeded in recovering SFEMS5 (SeaFloor ElectroMagnetic Station 5) by acoustic release on November 14th, 2016. SFEMS5 was deployed during the R/V Kairei cruise in May, 2013 (KR13-09), for the detection of tsunami-generated electromagnetic (EM) fields. All equipment attached to SFEMS5 was safely recovered without any glass sphere flooding. We obtained long-term EM time series over 2.8 years. The obtained long-term data will be useful not only to study tsunami-related EM fields but also to investigate the secular variation of the geomagnetic main field and couplings between EM fields with other oceanic flows with time scales longer than tsunamis.

1.3.3 Ocean depth sounding just after the off Fukushima Earthquake on Nov. 22nd, 2016

During the present cruise, a tsunamigenic earthquake (M7.4) occurred ~20 km off Fukushima prefecture at 05:59:58 JST on November 22nd, 2016. National Research Institute for Earth Science and Disaster Prevention rapidly estimated the location of the hypocenter as (141.3˚E, 37.3˚N, -11km). In the very vicinity of the focal area, we conducted a 3 mile × 3 mile acoustic survey of ocean depth by Multibeam Echo Sounder on November 23rd. We succeeded in obtaining dense ocean depth dataset with a horizontal spatial resolution of ~10 m as well as sea surface gravity and ship-board vector magnetic data. The obtained data can be compared with the past/future datasets in order to investigate the seafloor crustal deformation due to the tsunamigenic earthquake.

2. Response of marine ecosystem to the ocean acidification in the subarctic western North Pacific

2.1 CTD cast and water sampling

Masahide WAKITA (JAMSTEC): Principal investigator
Hiroshi Uchida (JAMSTEC)
Shungo OSHITANI (MWJ): Operation leader
Shinsuke TOYODA (MWJ)

(1) Objective

Investigation of oceanic structure and water sampling.

(2) Parameters

Temperature (Primary and Secondary)
Conductivity (Primary and Secondary)
Pressure
Dissolved Oxygen
Dissolved Oxygen voltage
Transmission % and beam attenuation coefficient and voltage
Fluorescence
Photosynthetically Active Radiation
Altimeter

(3) Instruments and Methods

CTD/Carousel Water Sampling System, which is a 12-position Carousel water sampler (CWS) with Sea-Bird Electronics, Inc. CTD (SBE9plus), was used during this cruise. 12-litter Niskin Bottles, which were washed by alkaline detergent and HCl, were used for sampling seawater. The sensors attached on the CTD were temperature (Primary and Secondary), conductivity (Primary and Secondary), pressure, dissolved oxygen, RINKO-III (dissolved oxygen sensor), dissolved oxygen (SBE43), transmission, fluorescence, PAR, altimeter and deep ocean standards thermometer. The Practical Salinity was calculated by measured values of pressure, conductivity and temperature. The CTD/CWS was deployed from starboard on working deck.

The CTD raw data were acquired on real time using the Seasave-Win32 (ver.7.23.2) provided by Sea-Bird Electronics, Inc. and stored on the hard disk of the personal computer. Seawater was sampled during the up cast by sending fire commands from the personal computer. We stayed for 1 minute at above 500 m layers before fire command to stabilize CTD. At deeper layer, we stayed for 30 seconds. 7 casts of CTD measurements were conducted (Table 2.1-1).

Data processing procedures and used utilities of SBE Data Processing-Win32 (ver.7.23.2) and SEASOFT were as follows:

(The process in order)

DATCNV: Convert the binary raw data to engineering unit data. DATCNV also extracts bottle information where scans were marked with the bottle confirm bit during acquisition. The duration was set to 4.4 seconds, and the offset was set to 0.0 seconds.

TCORP (original module): Corrected the pressure sensitivity of the primary temperature (SBE3) sensor.
S/N 031464: -8.94556e-008 (degC/dbar)

RINKOCOR (original module): Corrected of the hysteresis of RINK-III voltage.

RINKOCORROS (original module): Corrected of the hysteresis of RINKO-III voltage bottle data.

BOTTLESUM: Create a summary of the bottle data. The data were averaged over 4.4 seconds.

ALIGNCTD: Convert the time-sequence of sensor outputs into the pressure sequence to ensure that all calculations were made using measurements from the same parcel of water. Dissolved oxygen data are systematically delayed with respect to depth mainly because of the long time constant of the dissolved oxygen sensor and of an additional delay from the transit time of water in the pumped pluming line. This delay was compensated by 5 seconds advancing dissolved oxygen sensor (SBE43) output (dissolved oxygen voltage) relative to the temperature data. RINKO-III voltage (User polyminal 0 – 2) were advanced 1second, transmission data and transmission voltage were advanced 2 seconds

WILDEDIT: Mark extreme outliers in the data files. The first pass of WILDEDIT obtained an accurate estimate of the true standard deviation of the data. The data were read in blocks of 1000 scans. Data greater than 10 standard deviations were flagged. The second pass computed a standard deviation over the same 1000 scans excluding the flagged values. Values greater than 20 standard deviations were marked bad. This process was applied to pressure, depth, temperature, conductivity and dissolved oxygen voltage (SBE43).

CELLTM: Remove conductivity cell thermal mass effects from the measured conductivity. Typical values used were thermal anomaly amplitude alpha = 0.03 and the time constant 1/beta = 7.0.

FILTER: Perform a low pass filter on pressure with a time constant of 0.15 second. In order to produce

zero phase lag (no time shift) the filter runs forward first then backward

WFILTER: Perform a median filter to remove spikes in the fluorescence data, transmission data and voltage data. A median value was determined by 49 scans of the window.

SECTIONU (original module of SECTION): Select a time span of data based on scan number in order to reduce a file size. The minimum number was set to be the starting time when the CTD package was beneath the sea-surface after activation of the pump. The maximum number of was set to be the end time when the package came up from the surface.

LOOPEDIT: Mark scans where the CTD was moving less than the minimum velocity of 0.0 m/s (traveling backwards due to ship roll).

DESPIKE (original module): Remove spikes of the data. A median and mean absolute deviation was calculated in 1-dbar pressure bins for both down and up cast, excluding the flagged values. Values greater than 4 mean absolute deviations from the median were marked bad for each bin. This process was performed 2 times for temperature, conductivity, dissolved oxygen voltage (SBE43), RINKO-III voltage.

DERIVE: Compute dissolved oxygen (SBE43).

BINAVG: Average the data into 1-dbar pressure bins.

DERIVE: Compute the Practical Salinity, sigma-theta and potential temperature.

SPLIT: Separate the data from an input .cnv file into down cast and up cast files.

Configuration file: yk1616a.xmlcon

Specifications of the sensors are listed below.

CTD: SBE911plus CTD system

Under water unit:

SBE9plus (S/N 09P27443-0677, Sea-Bird Electronics, Inc.)

Pressure sensor: Digiquartz pressure sensor (S/N 79511)

Calibrated Date: 22 Apr 2016

Temperature sensors:

Primary: SBE03-04F (S/N 031464, Sea-Bird Electronics, Inc.)

Calibrated Date: 01 May. 2015

Secondary: SBE03-05F (S/N 030893, Sea-Bird Electronics, Inc.)

Calibrated Date: 13 Aug. 2015

Conductivity sensors:

Primary: SBE04C (S/N 043036, Sea-Bird Electronics, Inc.)

Calibrated Date: 06 May. 2015

Secondary: SBE04-04/0 (S/N 042240, Sea-Bird Electronics, Inc.)

Calibrated Date: 17 Sep. 2015

Dissolved Oxygen sensor:

SBE43 (S/N 430330, Sea-Bird Electronics, Inc.)

Calibrated Date: 10 May. 2016

RINK-III (S/N 0037 (1204), Alec Electronics Co. Ltd.)

Transmissonmeter:

C-Star (S/N CST-1363DR, WET Labs, Inc.)

Calibrated Date: 22 Jan. 2016

Fluorescence:

Chlorophyll Fluorometer (S/N 2936, Seapoint Sensors, Inc.)

Photosynthetically Active Radiation:

PAR sensor (S/N 0049, Satlantic Inc.)

Calibrated Date: 22 Jan. 2009

Altimeter:

Benthos PSA-916T (S/N 66557, Teledyne Benthos, Inc.)

Deep Ocean Standards Thermometer:

SBE35 (S/N 0022, Sea-Bird Electronics, Inc.)

Calibrated Date: 12 Nov 2016

Carousel water sampler:

SBE32 (S/N 3227443-0389, Sea-Bird Electronics, Inc.)

Deck unit: SBE11plus (S/N 11P9833-0344, Sea-Bird Electronics, Inc.)

(4) Preliminary Results

During this cruise, 7 casts of CTD observation were carried out. Date, time and locations of the CTD casts are listed in Table 2.1-1. During this cruise, we judged noise, spike or shift in the data of some cast. These were as follows.

KNTM01: Dissolved oxygen (SBE43)

down 2142-2144 dbar: spike

down 2317-2322 dbar: spike

down 2647-2650 dbar: spike

Transmissonmeter

down 1570-1575 dbar: spike

down 2908-2916 dbar: spike

(5) Data archive

These data obtained in this cruise will be submitted to the Data Management Group of JAMSTEC, and will be opened to the public via “Data Research System for Whole Cruise Information in JAMSTEC (DARWIN)” in JAMSTEC web site.

<http://www.godac.jamstec.go.jp/darwin/e>

Table 2.1-1YK16-16 CTD casttable

Stnnbr	Castno	Date(UTC)	Time(UTC)		BottomPosition		Depth	Wire Out	HT Above Bottom	Max Depth	Max Pressure	CTD Filename	Remark
		(mmddyy)	Start	End	Latitude	Longitude							
NWP	1	111416	00:18	01:04	41-02.59N	159-57.57E	-	1000.0	-	991.9	1002.0	NWPM01	Shimizu
KNT	1	111916	04:14	06:21	44-00.30N	154-59.34E	5352.1	3005.5	-	2958.9	3004.0	KNTM01	Routine
KNT	2	111916	07:56	08:35	43-59.88N	155-01.54E	5377.5	299.8	-	300.4	303.0	KNTM02	Routine
KNT	3	111916	15:24	15:50	43-59.69N	155-00.56E	5375.4	200.5	-	200.3	202.0	KNTM03	P.P
KNT	4	111916	16:46	17:04	43-59.50N	155-00.45E	5377.0	199.6	-	203.3	205.0	KNTM04	POM
KNT	5	111916	17:48	18:10	43-59.70N	155-00.81E	5381.2	201.7	-	202.3	204.0	KNTM05	POM
KNT	6	111916	18:54	19:18	44-00.05N	155-01.01E	5377.0	200.2	-	203.3	205.0	KNTM06	Yoshikawa

Routine: Routine sampling

P.P.: Primary Production POM: Particulate Organic Matter

2.2 Salinity measurement

Masahide Wakita (JAMSTEC)
Hiroshi Uchida (JAMSTEC)

(1) Objective

To measure bottle salinity obtained by CTD casts, bucket sampling.

(2) Methods

a. Salinity Sample Collection

Seawater samples were collected with 10 liter Niskin bottles and bucket. The salinity sample bottles of the 250ml brown grass bottles with screw caps were used for collecting the sample water. Each bottle was rinsed three times with the sample water, and was filled with sample water to the bottle shoulder. The salinity sample bottles were sealed with plastic inner caps and screw caps because we took into consideration the possibility of storage for about a month. These caps were rinsed three times with the sample water before use. The bottle was stored for less than 1 month on the laboratory before the salinity measurement. The number of samples is total of ~40 for CTD and Bucket

b. Instruments and Method

The salinity analysis on the laboratory was carried out using the salinometer (Model 8400B "AUTOSAL"; Guildline Instruments Ltd.) with an additional peristaltic-type intake pump (Ocean Scientific International, Ltd.). Digital thermometers (Model D617; Tateyama Kagaku Ind.) were used. The thermometer monitored the ambient temperature and the bath temperature of the salinometer.

The measurement system was almost the same as Aoyama et al. (2002). The salinometer was operated in the air-conditioned laboratory at a bath temperature of 24 deg C. The measurement for each sample was done with the double conductivity ratio and defined as the median of 60 readings of the salinometer. Data collection was started 5 seconds after filling the cell with the sample and it took about 15 seconds to collect 60 readings by a personal computer. Data were taken for the sixth and seventh filling of the cell after rinsing five times. In the case of the difference between the double conductivity ratio of these two fillings being smaller than 0.00002, the average value of the double conductivity ratio was used to calculate the bottle salinity with the algorithm for practical salinity scale, 1978 (UNESCO, 1981). If the difference was greater than or equal to 0.00003, an eighth filling of the cell was done. In

(3) Data Archive

All measured data will be submitted to JAMSTEC Data Management Office (DMO) and is currently under its control.

(4) Reference

Aoyama, M. , T. Joyce, T. Kawano and Y. Takatsuki: Standard seawater comparison up to P129. Deep-Sea Research, I, Vol. 49, 1103-1114, 2002

UNESCO : Tenth report of the Joint Panel on Oceanographic Tables and Standards. UNESCO Tech. Papers in Mar. Sci., 36, 25 pp., 1981

2.3 Dissolved oxygen

Masahide WAKITA (JAMSTEC)

(1) Objectives

Determination of dissolved oxygen in seawater by Winkler titration.

(2) Instruments and Methods

Following procedure is based on an analytical method, entitled by "Determination of dissolved oxygen in sea water by Winkler titration", in the WHP Operations and Methods (Dickson, 1996).

a. Instruments

Burette for sodium thiosulfate and potassium iodate;

APB-620 manufactured by Kyoto Electronic Co. Ltd. / 10 cm^3 of titration vessel

Detector;

Automatic photometric titrator (DOT-05) manufactured by Kimoto Electronic Co. Ltd.

b. Reagents

Pickling Reagent I: Manganese chloride solution (3 mol dm^{-3})

Pickling Reagent II:

Sodium hydroxide (8 mol dm^{-3}) / sodium iodide solution (4 mol dm^{-3})

Sulfuric acid solution (5 mol dm^{-3})

Sodium thiosulfate (0.025 mol dm^{-3})

Potassium iodide (0.001667 mol dm^{-3})

CSK standard of potassium iodide:

Lot DCE2131, Wako Pure Chemical Industries Ltd., 0.0100N

c. Sampling

Seawater samples were collected with Niskin bottle attached to the CTD-system and surface bucket sampler. Seawater for oxygen measurement was transferred from sampler to a volume calibrated flask (ca. 100 cm3). Three times volume of the flask of seawater was overflowed. Temperature was measured by digital thermometer during the overflowing. Then two reagent solutions (Reagent I and II) of 1.0 cm3 each were added immediately into the sample flask and the stopper was inserted carefully into the flask. The sample flask was then shaken vigorously to mix the contents and to disperse the precipitate finely throughout. After the precipitate has settled at least halfway down the flask, the flask was shaken again vigorously to disperse the precipitate. The sample flasks containing pickled samples were stored in a laboratory until they were titrated.

d. Sample measurement

At least two hours after the re-shaking, the pickled samples were measured on board. 1 cm3 sulfuric acid solution and a magnetic stirrer bar were added into the sample flask and stirring began. Samples were titrated by sodium thiosulfate solution whose morality was determined by potassium iodate solution. Temperature of sodium thiosulfate during titration was recorded by a digital thermometer. During this cruise, we measured dissolved oxygen concentration using 1 set of the titration apparatus. Dissolved oxygen concentration (μmol kg^{-1}) was calculated by sample temperature during seawater sampling, salinity of the CTD sensor, and titrated volume of sodium thiosulfate solution without the blank.

e. Standardization and determination of the blank

Concentration of sodium thiosulfate titrant was determined by potassium iodate solution. Pure potassium iodate was dried in an oven at 130°C. 1.7835 g potassium iodate weighed out accurately was dissolved in deionized water and diluted to final volume of 5 dm^3 in a calibrated volumetric flask (0.001667 mol dm^{-3}). 10 cm^3 of the standard potassium iodate solution was added to a flask using a volume-calibrated dispenser. Then 90 cm^3 of deionized water, 1 cm^3 of sulfuric acid solution, and 1.0 cm^3 of pickling reagent solution II and I were added into the flask in order. Amount of titrated volume of sodium thiosulfate (usually 5 times measurements average) gave the morality of sodium thiosulfate titrant.

The oxygen in the pickling reagents I (1.0 cm3) and II (1.0 cm3) was assumed to be 0.0017 ml (Murray et al., 1968). The blank due to other than oxygen was determined as follows. 1 cm^3 of the standard potassium iodate solution were added to flask using a calibrated dispenser. Then 100 cm3 of deionized water, 1 cm^3 of sulfuric acid solution, and 1 cm^3 of pickling reagent solution II and I each were added into the flask in order. The blank was determined by difference between the first (1 cm^3 of KIO3) titrated volume of the sodium thiosulfate and the second (1 cm^3 of KIO3) one. The results of 3 times blank determinations were averaged.

f. Repeatability of sample measurement

Replicate samples were taken at every CTD casts. Total amount of the replicate sample pairs of good measurement was 4. The standard deviation of the replicate measurement was 0.06 μmol kg^{-1} that was calculated by a procedure in Guide to best practices for ocean CO_2 measurements Chapter4 SOP23 Ver.3.0 (2007).

(4) Preliminary result

All measured data will be submitted to JAMSTEC Data Management Office (DMO) and is currently under its control.

(5) References

Dickson, A.G., Determination of dissolved oxygen in sea water by Winkler titration. (1996)

Dickson, A.G., Sabine, C.L. and Christian, J.R. (Eds.), Guide to best practices for ocean CO2 measurements. (2007)
Culberson, C.H., WHP Operations and Methods July-1991 “Dissolved Oxygen”, (1991)
Japan Meteorological Agency, Oceanographic research guidelines (Part 1). (1999)
KIMOTO electric CO. LTD., Automatic photometric titrator DOT-01 Instruction manual

2.4 Nutrients

Masahide WAKITA (JAMSTEC MIO)
Tomomi SONE (MWJ)

(1) Objectives

The objectives of nutrients analyses during the R/V Yokosuka YK16-16 cruise in the Western North Pacific Ocean is as follows:

- Describe the present status of nutrients concentration with excellent comparability.

(2) Parameters

The determinants are nitrate, nitrite, silicate, phosphate and ammonia in the North Pacific Ocean.

(3) Summary of nutrients analysis

We made 1 QuAAtro run for the water columns sample at 4 casts during YK16-16. The total amount of layers of the seawater sample reached to 37. We made duplicate measurement. The station location for nutrients measurement is shown in Figure 2.4.1.

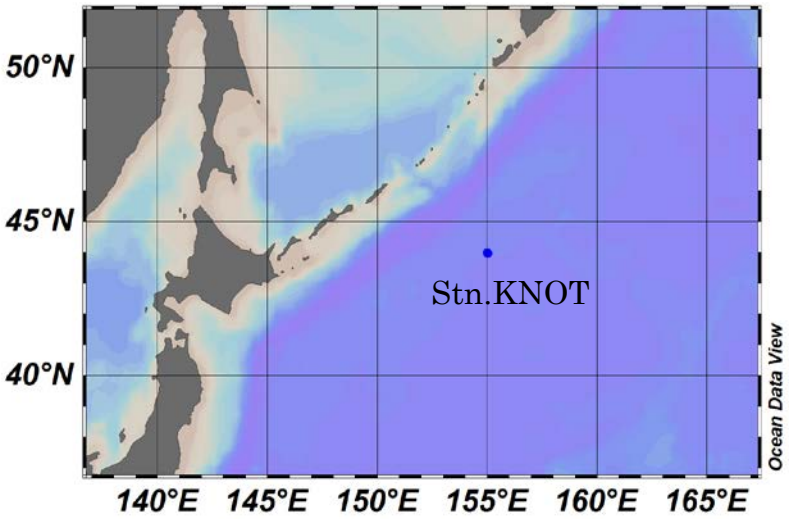


Figure 2.4.1. Sampling position of nutrients sample.

(4) Instrument and Method

(4.1) Analytical detail using QuAAtro 2-HR systems (BL-Tech)

Nitrate + nitrite and nitrite were analyzed according to the modification method of Grasshoff (1970).

The sample nitrate was reduced to nitrite in a cadmium tube inside of which was coated with metallic copper. The sample streamed with its equivalent nitrite was treated with an acidic, sulfanilamide reagent and the nitrite forms nitrous acid which reacted with the sulfanilamide to produce a diazonium ion. N-1-Naphthylethylene-diamine added to the sample stream then coupled with the diazonium ion to produce a red, azo dye. With reduction of the nitrate to nitrite, both nitrate and nitrite reacted and were measured; without reduction, only nitrite reacted. Thus, for the nitrite analysis, no reduction was performed and the alkaline buffer was not necessary. Nitrate was computed by difference.

The silicate method was analogous to that described for phosphate. The method used was essentially that of Grasshoff et al. (1983), wherein silicomolybdic acid was first formed from the silicate in the sample and added molybdic acid; then the silicomolybdic acid was reduced to silicomolybdous acid, or "molybdenum blue" using ascorbic acid as the reductant. The analytical methods of the nutrients, nitrate, nitrite, silicate and phosphate, during this cruise were same as the methods used in (Kawano et al. 2009).

The phosphate analysis was a modification of the procedure of Murphy and Riley (1962). Molybdic acid was added to the seawater sample to form phosphomolybdic acid which was in turn reduced to phosphomolybdous acid using L-ascorbic acid as the reductant.

The ammonia in seawater is mixed with an alkaline containing EDTA, ammonia as gas state is formed from seawater. The ammonia (gas) is absorbed in sulfuric acid by way of 0.5 μm pore size membrane filter (ADVANTEC PTFE) at the dialyzer attached to analytical system. The ammonia absorbed in sulfuric acid is determined by coupling with phenol and hypochlorite to form indophenols blue. Wavelength using ammonia analysis is 630 nm, which is absorbance of indophenols blue.

The details of modification of analytical methods used in this cruise are also compatible with the methods described in nutrients section in GO-SHIP repeat hydrography manual (Hydes et al., 2010). The flow diagrams and reagents for each parameter are shown in Figures 2.4.2. to 2.4.6.

(4.2) Nitrate + Nitrite Reagents

Imidazole (buffer), 0.06 M (0.4 % w/v)

Dissolve 4 g imidazole, $C_3H_4N_2$, in ca. 1000 ml DIW; add 2 ml concentrated HCl. After mixing, 1 ml Triton®X-100 (50 % solution in ethanol) is added.

Sulfanilamide, 0.06 M (1 % w/v) in 1.2M HCl

Dissolve 10 g sulfanilamide, 4-$NH_2C_6H_4SO_3H$, in 900 ml of DIW, add 100 ml concentrated HCl. After mixing, 2 ml Triton®X-100 (50 % solution in ethanol) is added.

N-1-Napthylethylene-diamine dihydrochloride, 0.004 M (0.1 %f w/v)

Dissolve 1 g NED, $C_{10}H_7NHCH_2CH_2NH_2$•2HCl, in 1000 ml of DIW and add 10 ml concentrated HCl. After mixing, 1 ml Triton®X-100 (50 % solution in ethanol) is added. This reagent is stored in a dark bottle.

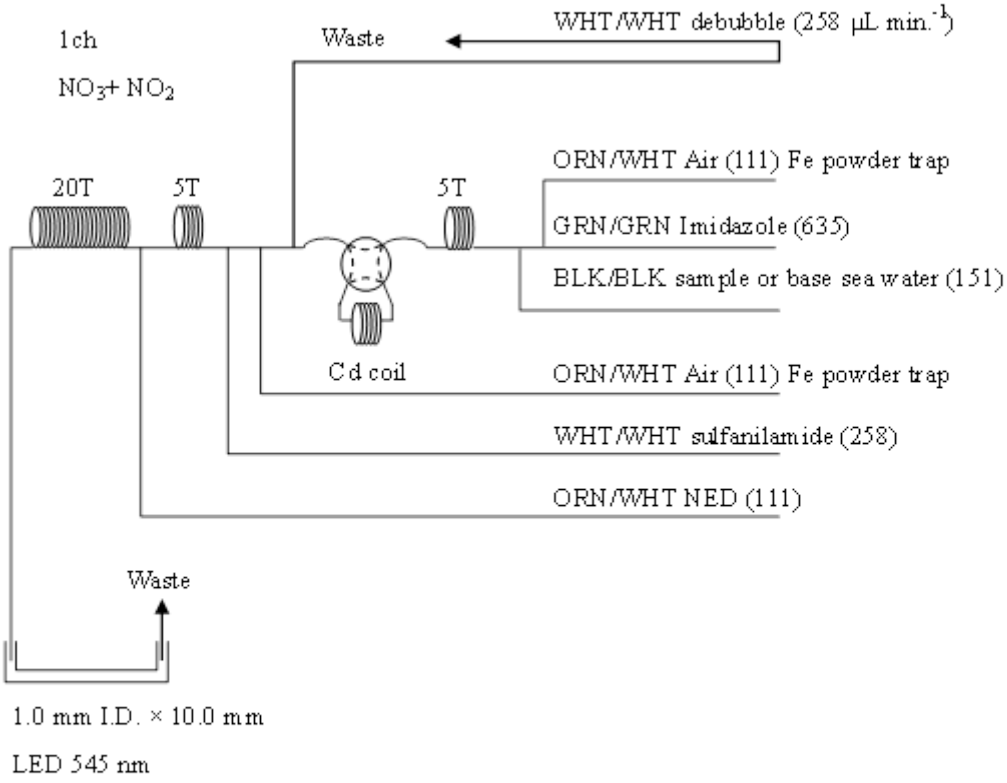


Figure 2.4.2. $NO_3 + NO_2$ (1ch.) Flow diagram.

(4.3) Nitrite Reagents

Sulfanilamide, 0.06 M (1 % w/v) in 1.2 M HCl

Dissolve 10g sulfanilamide, 4-$NH_2C_6H_4SO_3H$, in 900 ml of DIW, add 100 ml concentrated HCl. After mixing, 2 ml Triton®X-100 (50 % solution in ethanol) is added.

N-1-Napthylethylene-diamine dihydrochloride, 0.004 M (0.1 % w/v)

Dissolve 1 g NED, $C_{10}H_7NHCH_2CH_2NH_2 \cdot 2HCl$, in 1000 ml of DIW and add 10 ml concentrated HCl. After mixing, 1 ml Triton®X-100 (50 % solution in ethanol) is added. This reagent is stored in a dark bottle.

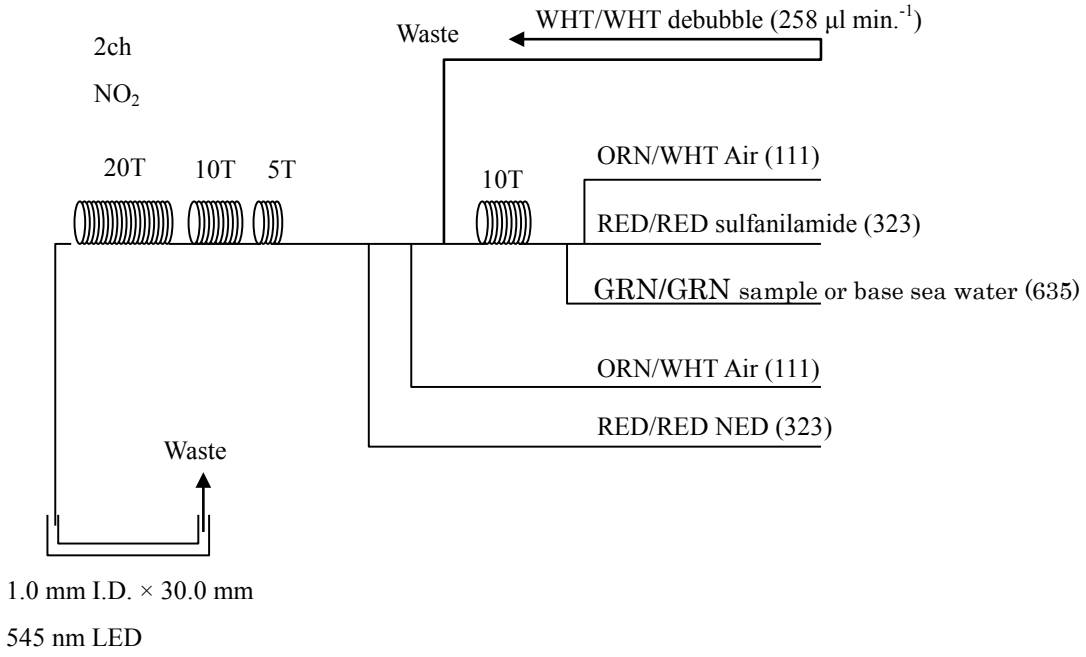


Figure2.4.3. NO_2 (2ch.) Flow diagram.

(4.4) Silicate Reagents

Molybdic acid, 0.06 M (2 % w/v)

Dissolve 15 g disodium molybdate(VI) dihydrate, $Na_2M_oO_4 \cdot 2H_2O$, in 980 ml DIW, add 8 ml concentrated H_2SO_4. After mixing, 20 ml sodium dodecyl sulphate (15 % solution in water) is added.

Oxalic acid, 0.6 M (5 % w/v)

Dissolve 50 g oxalic acid anhydrous, HOOC: COOH, in 950 ml of DIW.

Ascorbic acid, 0.01M (3 % w/v)

Dissolve 2.5g L (+)-ascorbic acid, $C_6H_8O_6$, in 100 ml of DIW. Stored in a dark bottle and freshly prepared before every measurement.

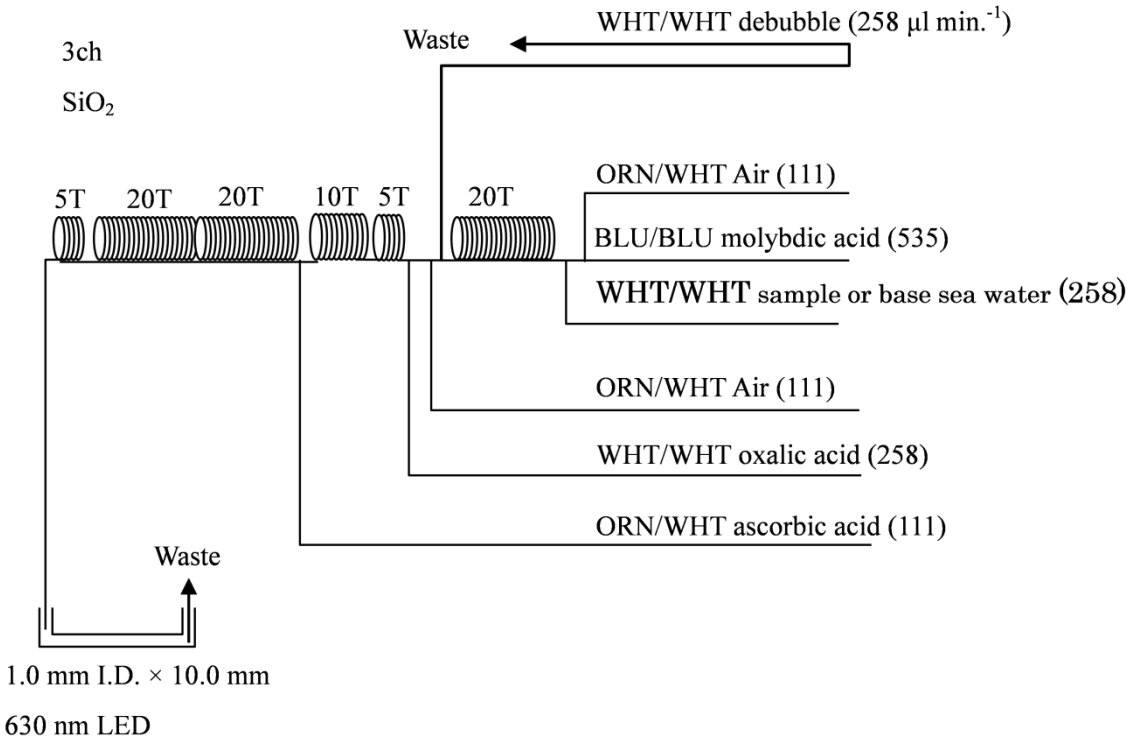


Figure2.4.4. SiO_2 (3ch.) Flow diagram.

(4.5) Phosphate Reagents

Stock molybdate solution, 0.03M (0.8 % w/v)

Dissolve 8 g disodium molybdate(VI) dihydrate, $Na_2M_oO_4 \bullet 2H_2O$, and 0.17 g antimony potassium tartrate, $C_8H_4K_2O_{12}Sb_2 \bullet 3H_2O$, in 950 ml of DIW and add 50 ml concentrated H_2SO_4.

Mixed Reagent

Dissolve 1.2 g L (+)-ascorbic acid, $C_6H_8O_6$, in 150 ml of stock molybdate solution. After mixing, 3 ml sodium dodecyl sulphate (15 % solution in water) is added. Stored in a dark bottle and freshly prepared before every measurement.

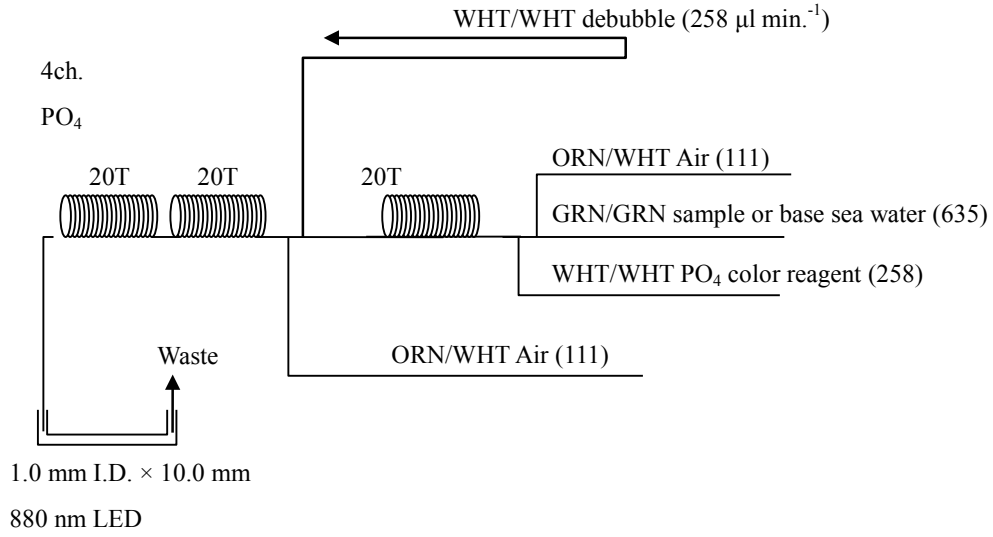


Figure2.4.5. PO_4 (4ch.) Flow diagram.

(4.6) Ammonia Reagents

EDTA

Dissolve 41 g EDTA (ethylenediaminetetraacetatic acid tetrasodium salt), $C_{10}H_{12}N_2O_8Na_4 \bullet 4H_2O$, and 2 g boric acid, H_3BO_3, in 200 ml of DIW. After mixing, 1 ml Triton®X-100 (30 % solution in DIW) is added. This reagent is prepared at a week about.

NaOH

Dissolve 5 g sodium hydroxide, NaOH, and 16 g EDTA in 100 ml of DIW. This reagent is prepared at a week about.

Stock Nitroprusside

Dissolved 0.25 g sodium pentacyanonitrosylferrate(II), $Na_2[Fe(CN)_5NO]$, in 100 ml of DIW and add 0.2 ml 1N H_2SO_4. Stored in a dark bottle and prepared at a month about.

Nitroprusside solution

Mixed 4 ml stock nitroprusside and 5 ml 1N H_2SO_4 in 500 ml of DIW. After mixing, 2ml Triton®X-100 (30 % solution in DIW) is added. This reagent is stored in a dark bottle and prepared at every 2 or 3 days.

Alkaline phenol

Dissolved 10 g phenol, C_6H_5OH, 5 g sodium hydroxide and citric acid, $C_6H_8O_7$, in 200 ml DIW. Stored in a dark bottle and prepared at a week about.

NaClO solution

Mixed 3 ml sodium hypochlorite solution, NaClO, in 47 ml DIW. Stored in a dark bottle and fleshly prepared before every measurement. This reagent is prepared 0.3% available chlorine.

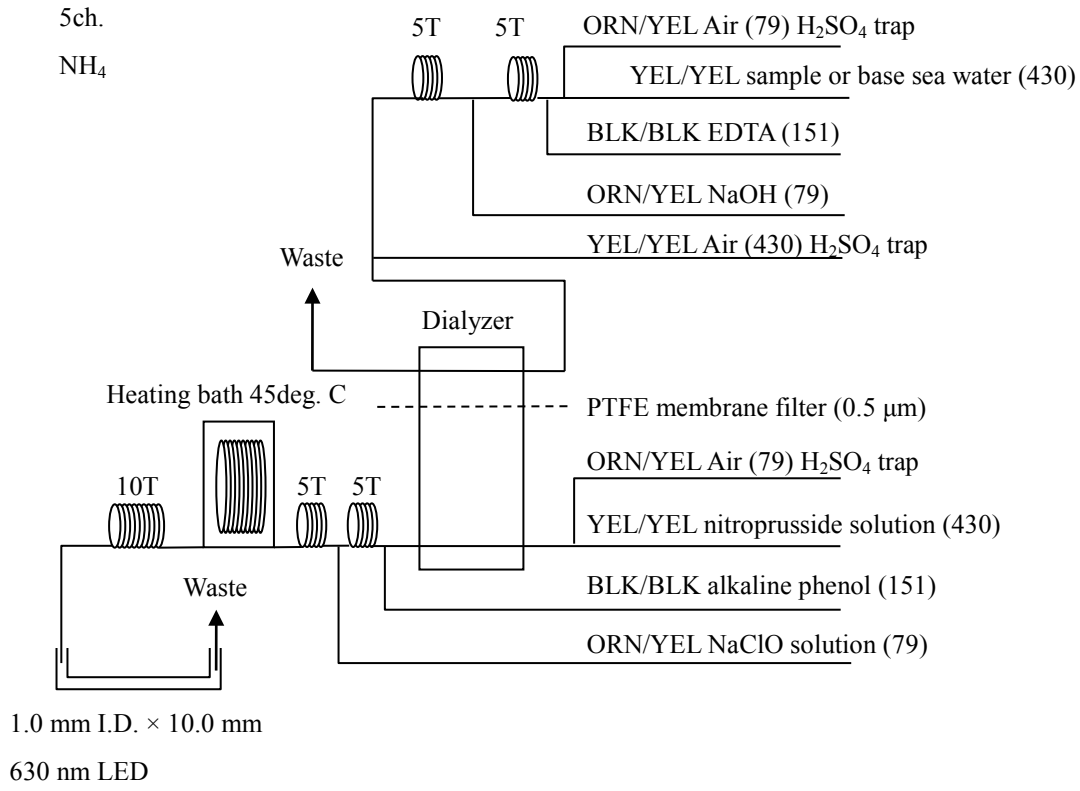


Figure 2.4.6 NH_4 (5ch.) Flow diagram.

(4.7) Sampling procedures

Sampling of nutrients followed that oxygen, salinity and trace gases. Samples were drawn into a virgin 10 ml polyacrylates vials without sample drawing tubes. These were rinsed three times before filling and vials were capped immediately after the drawing. The vials ware kept to ambient temperature, 20.0 ± 1.0 deg. C, in about 60 minutes before use to stabilize the temperature of samples.

No transfer was made and the vials were set an auto sampler tray directly. Samples were analyzed within 24 hours after collection.

(4.8) Data processing

Raw data from QuAAtro were treated as follows:

- Check baseline shift.
- Check the shape of each peak and positions of peak values taken, and then change the positions of peak values taken if necessary.
- Carry-over correction and baseline drift correction were applied to peak heights of each samples followed

by sensitivity correction.

- Baseline correction and sensitivity correction were done basically using liner regression.
- Calibration curves to get nutrients concentration were assumed second order equations.

(5) Nutrients standards

(5.1) Volumetric laboratory ware of in-house standards

All volumetric glass ware and polymethylpentene (PMP) ware used were gravimetrically calibrated. Plastic volumetric flasks were gravimetrically calibrated at the temperature of use within 4 K.

(5.1.1) Volumetric flasks

Volumetric flasks of Class quality (Class A) are used because their nominal tolerances are 0.05 % or less over the size ranges likely to be used in this work. Class A flasks are made of borosilicate glass, and the standard solutions were transferred to plastic bottles as quickly as possible after they are made up to volume and well mixed in order to prevent excessive dissolution of silicate from the glass. PMP volumetric flasks were gravimetrically calibrated and used only within 4 K of the calibration temperature.

The computation of volume contained by glass flasks at various temperatures other than the calibration temperatures were done by using the coefficient of linear expansion of borosilicate crown glass.

Because of their larger temperature coefficients of cubical expansion and lack of tables constructed for these materials, the plastic volumetric flasks were gravimetrically calibrated over the temperature range of intended use and used at the temperature of calibration within 4 K. The weights obtained in the calibration weightings were corrected for the density of water and air buoyancy.

(5.1.2) Pipettor

The pipettor used in this cruise has nominal calibration tolerances of 0.1 % or better. It was gravimetrically calibrated in order to verify and improve upon this nominal tolerance.

(5.2) Reagents, general considerations

(5.2.1) Specifications

For nitrate standard, “potassium nitrate 99.995 suprapur®” provided by Merck, Lot. B0771365211, CAS No.: 7757-91-1, was used.

For nitrite standard solution, we used “nitrous acid iron standard solution (NO_2^- 1000) provided by Wako, Lot ECP4122, Code. No. 140-06451.” This standard solution was certified by Wako using Ion chromatograph method. Calibration result is 999 mg L^{-1} at 20 degree Celsius. Expanded uncertainty of calibration (k=2) is 0.7 % for the calibration result.

For phosphate standard, “potassium dihydrogen phosphate anhydrous 99.995 suprapur®” provided by Merck, Lot. B1144508528, CAS No.: 7778-77-0, was used.

For the silicate standard, we use “Silicon standard solution SiO_2 in NaOH 0.5 mol/l CertiPUR®” provided by Merck, CAS No.: 1310-73-2, of which lot number is HC54715536 are used. The silicate concentration is certified by NIST-SRM3150 with the uncertainty of 0.7 %. HC54715536 is certified as 1005 mg L^{-1}.

For ammonia standard, “ammonium Chloride” provided by NMIJ. We used NMIJ CRM 3011-a. The purity of this standard was greater than 99.9 %. Expanded uncertainty of calibration (k=2) is 0.065 %.

(5.2.2) Ultra-pure water

Ultra-pure water (Milli-Q water) freshly drawn was used for preparation of reagent, standard solutions and for measurement of reagent and system blanks.

(5.2.3) Low nutrients seawater (LNSW)

Surface water having low nutrient concentration was taken and filtered using 0.20 μm pore capsule cartridge filter at MR15-05 cruise on January, 2016. This water is stored in 20 liter cubitainer with paper box.

LNSW concentrations were assigned to August, 2016 on MR16-06 cruise.

(5.3) Concentrations of nutrients for A, B and C standards

Concentrations of nutrients for A, B and C standards are set as shown in Table 2.4.1. Then the actual concentration of nutrients in each fresh standard was calculated based on the ambient, solution temperature and determined factors of volumetric laboratory wares.

The calibration curves for each run were obtained using 5 levels, C-1, C-2, C-3, C-4 and C-5 for nitrate, nitrite, silicate and phosphate and using 4 levels, C-1, C-3, C-4 and C-5 for ammonia.

Table 2.4.1. Nominal concentrations of nutrients for A, B and C.

	A	B	C-1	C-2	C-3	C-4	C-5
NO_3 (μM)	23000	900	0	9	18	36	54
NO_2 (μM)	21700	20	0	0.2	0.4	0.9	1.3
SiO_2 (μM)	36000	2850	0.8	29	58	115	172
PO_4 (μM)	3000	60	0.01	0.6	1.2	2.4	3.6
NH_4(μM)	4000	120	0	—	1.2	2.4	3.6

Table 2.4.2. Working calibration standard recipes.

C Std.	B-1 Std.	B-2 Std.	B-3 Std.	DIW

C-1	0 ml	0 ml	0 ml	75 ml
C-2	5 ml	5 ml	—	65 ml
C-3	10 ml	10 ml	5 ml	50 ml
C-4	20 ml	20 ml	10 ml	25 ml
C-5	30 ml	30 ml	15 ml	0 ml

B-1 Std.: Mixture of nitrate, silicate and phosphate

B-2 Std.: Nitrite

B-3 Std.: Ammonia

(5.4) Renewal of in-house standard solutions

In-house standard solutions were renewed as shown in Table 2.4.3(a), (b) and (c).

Table 2.4.3(a). Timing of renewal of in-house standards.

NO_3, NO_2, SiO_2, PO_4, NH_4	Renewal
A-1 Std. (NO_3)	maximum a month
A-2 Std. (NO_2)	commercial prepared solution
A-3 Std. (SiO_2)	commercial prepared solution
A-4 Std. (PO_4)	maximum a month
A-5 Std. (NH_4)	maximum a month
D-1 Std.	maximum 8 days
D-2 Std.	maximum 8 days
B-1 Std. (mixture of NO_3, SiO_2, PO_4)	maximum 8 days
B-2 Std. (NO_2)	maximum 8 days
B-3 Std. (NH_4)	maximum 8 days

Table 2.4.3(b) Timing of renewal of working calibration standards.

C Std.	Renewal
C Std. (mixture of B-1, B-2 and B-3 Std.)	every 24 hours

Table 2.4.3(c) Timing of renewal of in-house standards for reduction estimation.

Reduction estimation	Renewal
45 μM NO_3	when C Std. renewed

43 μM NO_2		when C Std. renewed

(6) Certified Reference Material of nutrients in seawater

To get the more accurate and high quality nutrients data to achieve the objectives stated above, huge numbers of the bottles of the Certified Reference Material of nutrients in seawater (hereafter CRM), which were recently certify by JAMSTEC and KANSO, are prepared (Aoyama et al., 2006, 2007, 2008, 2009, 2012, 2014, 2016). In the previous worldwide expeditions, such as WOCE cruises, the higher reproducibility and precision of nutrients measurements were required (Joyce and Corry, 1994). Since no standards were available for the measurement of nutrients in seawater at that time, the requirements were described in term of reproducibility. The required reproducibility was 1 %, 1 to 2 %, 1 to 3 % for nitrate, phosphate and silicate, respectively. Although nutrient data from the WOCE one-time survey was of unprecedented quality and coverage due to much care in sampling and measurements, the differences of nutrients concentration at crossover points are still found among the expeditions (Aoyama and Joyce, 1996, Mordy et al., 2000, Gouretski and Jancke, 2001). For instance, the mean offset of nitrate concentration at deep waters was 0.5 μmol kg^{-1} for 345 crossovers at world oceans, though the maximum was 1.7 μmol kg^{-1} (Gouretski and Jancke, 2001). At the 31 crossover points in the Pacific WHP one-time lines, the WOCE standard of reproducibility for nitrate of 1 % was fulfilled at about half of the crossover points and the maximum difference was 7 % at deeper layers below 1.6 degree Celsius in potential temperature (Aoyama and Joyce, 1996).

(6.1) CRM for this cruise

RMNS lots BY, BU, BW, BV and BZ, which cover full range of nutrients concentrations in the North Pacific ocean are prepared.

(6.2) CRM concentration

We used nutrients concentrations for CRM lots BY, BU, BW, BV and BZ as shown in Table 2.4.4.

Table 2.4.4 Certified concentration and uncertainty (k=2) of CRMs.

unit: μmol kg^{-1}

Lot	Nitrate	Nitrite	Silicate	Phosphate	Ammonia*
BY	0.02 ± 0.02	0.02 ± 0.01	1.76 ± 0.06	0.039 ± 0.010	0.90
BU	3.94 ± 0.05	0.07 ± 0.01	20.92 ± 0.49	0.345 ± 0.009	0.99
BW	24.59 ± 0.20	0.07 ± 0.01	60.01 ± 0.42	1.541 ± 0.014	0.93
BV	35.36 ± 0.35	0.05 ± 0.01	102.2 ± 1.10	2.498 ± 0.023	1.02
BZ	43.35 ± 0.33	0.22 ± 0.01	161.0 ± 0.93	3.056 ± 0.033	0.44

*For ammonia values are references

(7) Quality control

(7.1) Precision of nutrients analyses during the measurement

Precision of nutrients analyses during this cruise was evaluated based on the repeated measurement of C-5 std, which are measured every 8 to 14 samples during the run. The analytical precisions are shown as shown in Table 2.4.5.

Analytical precisions previously evaluated were 0.08 % for nitrate, 0.07 % for silicate and 0.10 % for phosphate in CLIVAR P21 revisited cruise of MR09-01 cruise in 2009, respectively. In this cruise, analytical precisions were 0.17% for nitrate, 0.26% for nitrite, 0.13% for silicate, 0.14% for phosphate and 0.61% for ammonia.

Table 2.4.5 Analytical precision based on the repeated measurement of C-5 std.

	Nitrate CV %	Nitrite CV %	Silicate CV %	Phosphate CV %	Ammonia CV %
analytical precision	0.17	0.26	0.13	0.14	0.61

(7.2) Carry over

The magnitudes of carry over throughout the measurement are shown in Table 2.4.6. These are small enough within acceptable levels.

Table 2.4.6. Carry over throughout the measurement.

	Nitrate %	Nitrite %	Silicate %	Phosphate %	Ammonia %
carry over	0.14	0.13	0.13	0.12	0.12

(8) Data archive

These data obtained in this cruise will be submitted to JAMSTEC.

(9) References

Aminot, A. and Kerouel, R. 1991. Autoclaved seawater as a reference material for the determination of nitrate and phosphate in seawater. Anal. Chim. Acta, 248: 277-283.

Aminot, A. and Kirkwood, D.S. 1995. Report on the results of the fifth ICES intercomparison exercise for nutrients in sea water, ICES coop. Res. Rep. Ser., 213.

Aminot, A. and Kerouel, R. 1995. Reference material for nutrients in seawater: stability of nitrate, nitrite,

ammonia and phosphate in autoclaved samples. Mar. Chem., 49: 221-232.

Aoyama M., and Joyce T.M. 1996, WHP property comparisons from crossing lines in North Pacific. In Abstracts, 1996 WOCE Pacific Workshop, Newport Beach, California.

Aoyama, M., 2006: 2003 Intercomparison Exercise for Reference Material for Nutrients in Seawater in a Seawater Matrix, Technical Reports of the Meteorological Research Institute No.50, 91pp, Tsukuba, Japan.

Aoyama, M., Susan B., Minhan, D., Hideshi, D., Louis, I. G., Kasai, H., Roger, K., Nurit, K., Doug, M., Murata, A., Nagai, N., Ogawa, H., Ota, H., Saito, H., Saito, K., Shimizu, T., Takano, H., Tsuda, A., Yokouchi, K., and Agnes, Y. 2007. Recent Comparability of Oceanographic Nutrients Data: Results of a 2003 Intercomparison Exercise Using Reference Materials. Analytical Sciences, 23: 1151-1154.

Aoyama M., J. Barwell-Clarke, S. Becker, M. Blum, Braga E. S., S. C. Coverly,E. Czobik, I. Dahllof, M. H. Dai, G. O. Donnell, C. Engelke, G. C. Gong, Gi-Hoon Hong, D. J. Hydes, M. M. Jin, H. Kasai, R. Kerouel, Y. Kiyomono, M. Knockaert, N. Kress, K. A. Krogslund, M. Kumagai, S. Leterme, Yarong Li, S. Masuda, T. Miyao, T. Moutin, A. Murata, N. Nagai, G.Nausch, M. K. Ngirchechol, A. Nybakk, H. Ogawa, J. van Ooijen, H. Ota, J. M. Pan, C. Payne, O. Pierre-Duplessix, M. Pujo-Pay, T. Raabe, K. Saito, K. Sato, C. Schmidt, M. Schuett, T. M. Shammon, J. Sun, T. Tanhua, L. White, E.M.S. Woodward, P. Worsfold, P. Yeats, T. Yoshimura, A.Youenou, J. Z. Zhang, 2008: 2006 Intercomparison Exercise for Reference Material for Nutrients in Seawater in a Seawater Matrix, Technical Reports of the Meteorological Research Institute No. 58, 104pp.

Gouretski, V.V. and Jancke, K. 2001. Systematic errors as the cause for an apparent deep water property variability: global analysis of the WOCE and historical hydrographic data・REVIEW ARTICLE, Progress In Oceanography, 48: Issue 4, 337-402.

Grasshoff, K., Ehrhardt, M., Kremling K. et al. 1983. Methods of seawater anylysis. 2nd rev. Weinheim: Verlag Chemie, Germany, West.

Joyce, T. and Corry, C. 1994. Requirements for WOCE hydrographic programmed data reporting. WHPO Publication, 90-1, Revision 2, WOCE Report No. 67/91.

Kawano, T., Uchida, H. and Doi, T. WHP P01, P14 REVISIT DATA BOOK, (Ryoin Co., Ltd., Yokohama, 2009).

Kirkwood, D.S. 1992. Stability of solutions of nutrient salts during storage. Mar. Chem., 38 : 151-164.

Kirkwood, D.S. Aminot, A. and Perttila, M. 1991. Report on the results of the ICES fourth intercomparison exercise for nutrients in sea water. ICES coop. Res. Rep. Ser., 174.

Mordy, C.W., Aoyama, M., Gordon, L.I., Johnson, G.C., Key, R.M., Ross, A.A., Jennings, J.C. and Wilson. J. 2000. Deep water comparison studies of the Pacific WOCE nutrient data set. Eos Trans-American Geophysical Union. 80 (supplement), OS43.

Murphy, J., and Riley, J.P. 1962. Analytica chim. Acta 27, 31-36.

Uchida, H. & Fukasawa, M. WHP P6, A10, I3/I4 REVISIT DATA BOOK Blue Earth Global Expedition 2003 1, 2, (Aiwa Printing Co., Ltd., Tokyo, 2005).

2.5 Dissolved inorganic carbon and Total alkalinity

Masahide WAKITA (JAMSTEC)

(1) Purpose of the study

Concentration of CO_2 in the atmosphere is now increasing owing to human activities such as burning of fossil fuels, deforestation, and cement production. The ocean plays an important role in buffering the increase of atmospheric CO_2. Anthropogenic CO_2 emitted into the atmosphere as a result of human activities was globally taken up by oceans at a rate of 2.2 ± 0.4 Pg C y–1 during the 1990s [IPCC, 2007]. Ocean acidification is a direct consequence of the ocean absorbing large amounts of the anthropogenic CO_2. The CO_2 uptake by the oceans has led to lowering both pH and $CaCO_3$ saturation states with regard to the mineral phases due to increasing hydrogen ions (H^+) and declining carbonate ion (CO_3^{2-}), respectively. Because oceanic biological activity has an important role concerned to Carbon cycle in the ocean through its photosynthesis and respiration, the chemical changes associated with ocean acidification have the potential to affect ocean biogeochemistry and ecosystems in a myriad of ways. Therefore, it is important to clarify the mechanism of the oceanic CO_2 absorption and ocean acidification and to estimate CO_2 absorption capacity and decrease of pH and $CaCO_3$ saturation states in recent years. When CO_2 dissolves in water, chemical reaction takes place and CO_2 alters its appearance into several species. Concentrations of the individual species of the CO_2 system in solution cannot be measured directly, but calculated from two of four parameters: total dissolved inorganic carbon (DIC), total alkalinity (TA), pH and pCO_2. This study presents the distribution of DIC and TA in the North Pacific Ocean.

(2) Sampling

Seawater samples of DIC and TA were collected by 10 liter Niskin bottles mounted on the CTD/Carousel Water Sampling System and a bucket at K2 station and brought the total to ~40. Seawaters were sampled in a 150 ml glass bottle for DIC and a 100 ml glass bottle for TA. These bottles were previously soaked in 1M HCl solution at least 6 hours and was cleaned by fresh water for 7 times and Milli-Q deionized water for 3 times. A sampling silicone rubber tube with PFA tip was connected to the Niskin bottle when the sampling was carried out. The glass bottles were filled from the bottom, without rinsing, and were overflowed for 20 seconds. After collecting the samples on the deck, the glass bottles were carried to the laboratory. Within one hour after the sampling, 1 % by the bottle volume (1 ml) was removed from the glass bottle and poisoned with 0.05% by volume (0.05 ml) of over saturated solution of mercury chloride. Then, the samples were sealed by rubber and aluminum caps. All samples preserved at ~ 5°C cold until analysis.

(3) Analysis

DIC and TA samples were measured by using coulometric and potentiometric techniques,

respectively, according to Dickson et al., 2007. The DIC and TA values will be determined with calibration against certified reference material provided by Prof. A. G. Dickson (Scripps Institution of Oceanography) and KANSO.

(4) Preliminary result

The distributions of DIC and TA will be determined as soon as possible after this cruise.

2.6 Dissolved organic carbon (DOC) and Total dissolved nitrogen (TDN)

Masahide WAKITA (JAMSTEC MIO)

(1) Purpose of the study

Variabilities in the concentration of dissolved organic carbon (DOC) in seawater have a potentially great impact on the carbon cycle in the marine system, because DOC is a major global carbon reservoir. A change by < 10% in the size of the oceanic DOC pool, estimated to be ~ 700 GtC (IPCC, 2007), would be comparable to the annual primary productivity in the whole ocean. In fact, it was generally concluded that the bulk DOC in oceanic water, especially in the deep ocean, is quite inert based upon ^{14}C-age measurements. Nevertheless, it is widely observed that in the ocean DOC accumulates in surface waters at levels above the more constant concentration in deep water, suggesting the presence of DOC associated with biological production in the surface ocean. This study presents the distribution of DOC in the North Pacific.

(2) Sampling

Seawater samples of DOC and TDN were collected by 10 liter Niskin bottles mounted on the CTD/Carousel Water Sampling System and a bucket at K2 stations and brought the total to ~100. Seawater from each Niskin bottle was transferred into 60 ml High Density Polyethylene bottle (HDPE) rinsed with same water three times. Water taken from the surface to bottom is filtered using precombusted (450°C) GF/F inline filters as they are being collected from the Niskin bottle. After collection, samples are frozen upright and preserved at ~ -20 °C cold until analysis in our land laboratory. Before use, all glassware was muffled at 550 °C for 5 hrs.

(3) Analysis

Prior to analysis, samples are returned to room temperature and acidified to pH < 2 with concentrated hydrochloric acid. DOC/TDN analysis was basically made with a high-temperature catalytic oxidation (HTCO) system improved a commercial unit, the Shimadzu TOC-L (Shimadzu Co.). In this system, the non-dispersive infrared was used for carbon dioxide produced from DOC during the HTCO process (temperature: 680 °C, catalyst: 0.5% Pt-Al_2O_3). Non-purgeable dissolved nitrogen compounds are combusted and converted to NO which, when mixed with ozone, chemiluminesces for detection by a photomultiplier

(4) Preliminary result

The distributions of DOC and TDN will be determined as soon as possible after this cruise.

2.7 Particle organic matters

Yoshihisa MINO (Nagoya University)

(1) Objective

Carbon and nitrogen stable isotope ratios ($\delta^{13}C$ and $\delta^{15}N$) of particulate organic matters in the ocean can provide insights into biogeochemical processes, formation and microbial transformation of particles since a mass-dependent isotopic fractionation occurs in each pathway. In this study we examined the vertical distribution of $\delta^{13}C$ and $\delta^{15}N$ of suspended particles to elucidate particle dynamics in the upper ocean of the western subarctic North Pacific in autumn when the mixed layer deepens.

(2) Sampling

About 20 to 40 liters of seawater were collected by CTD-RMS at the depths from surface to 200 m depths, and filtered through pre-combusted GF/F filters (Whatman) and the filters were kept frozen until analysis on shore.

(3) Analysis

The filter samples of suspended particles are exposed to HCl fumes overnight to remove carbonates, dried in vacuum, and then pelletized with a tin disk. Amount of particulate organic carbon and nitrogen (POC, PN) and both isotopes of particles in the pellets are measured with an elemental analyzer combined with a continuous flow isotope-ratio mass spectrometer (EA1112-Delta Plus, Thermo Fisher Scientific).

(4) Preliminary result

The distributions of $\delta^{13}C$ and $\delta^{15}N$ as well as POC and PN concentrations of suspended particles will be determined as soon as possible after this cruise.

(5) Data archive

All data will be submitted to JAMSTEC Data Management Group (DMG) within 2 years.

2.8 Phytoplankton

2.8.1 Chlorophyll *a* measurements by fluorometric determination

Tetsuichi FUJIKI (JAMSTEC) : Principal Investigator
Keitaro MATSUMOTO (MWJ) : Operation Leader

1. Objective

Phytoplankton biomass can estimate as the concentration of chlorophyll *a* (chl-*a*), because all oxygenic photosynthetic plankton contain chl-*a*. Phytoplankton exist various species in the ocean, but the species are roughly characterized by their cell size. The objective of this study is to investigate the vertical distribution of phytoplankton and their size fractionations as chl-*a* by using the fluorometric determination.

2. Sampling

Samplings of total chl-*a* were conducted from 13 depths between the surface and 300 m. At the cast for primary production, water samples were collected 7 depths in the euphotic layer at the station of KNOT.

3. Instruments and Methods

Water samples (0.5L) for total chl-*a* were filtered (<0.02 MPa) through 25mm-diameter Whatman GF/F filter. Size-fractionated chl-*a* were obtained by sequential filtration (<0.02 MPa) of 1-L water sample through 10-µm, 3-µm and 1-µm polycarbonate filters (47-mm diameter) and Whatman GF/F filter (25-mm diameter). Phytoplankton pigments retained on the filters were immediately extracted in a polypropylene tube with 7 ml of N,N-dimethylformamide (Suzuki and Ishimaru, 1990). Those tubes were stored at –20°C under the dark condition to extract chl-*a* for 24 hours or more.

Fluorescences of each sample were measured by Turner Design fluorometer (10-AU-005), which was calibrated against a pure chl-*a* (Sigma-Aldrich Co.). We applied two kind of fluorometric determination for the samples of total chl-*a*: "Non-acidification method" (Welschmeyer, 1994). Size-fractionated samples were applied only "Non-acidification method". Analytical conditions of each method were listed in table 2.9.1-1.

4. Preliminary Results

The results of total chl-*a* at station KNOT were shown in Figure 2.8.1-1. The results of size fractionated chl-*a* were shown in Figure2.8.1-2.

5. Data archives

The processed data file of pigments will be submitted to the JAMSTEC Data Management Group (DMG) within a restricted period. Please ask PI for the latest information.

6. Reference

Suzuki, R., and T. Ishimaru (1990), An improved method for the determination of phytoplankton chlorophyll using N, N-dimethylformamide, *J. Oceanogr. Soc. Japan*, 46, 190-194.

Holm-Hansen, O., Lorenzen, C. J., Holmes, R.W. and J. D. H. Strickland (1965), Fluorometric determination of chlorophyll. *J. Cons. Cons. Int. Explor. Mer.* 30, 3-15.

Welschmeyer, N. A. (1994), Fluorometric analysis of chlrophyll *a* in the presence of chlorophyll *b* and pheopigments. *Limnol. Oceanogr.* 39, 1985-1992.

Table 2.8.1. Analytical conditions of "Non-acidification method" for chlorophyll *a* with Turner Designs fluorometer (10-AU-005).

Excitation filter	436 nm
Emission filter	680 nm
Lamp	Blue Mercury Vapor

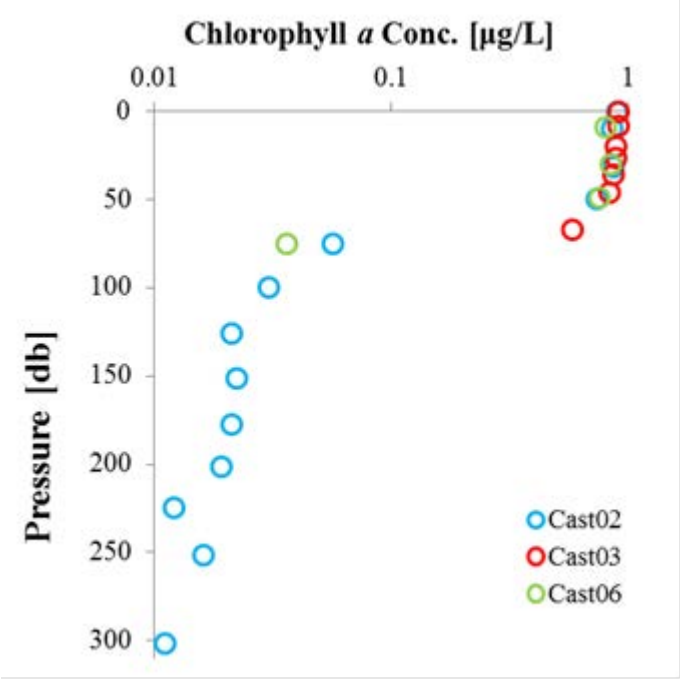


Figure 2.8.1-1 Vertical distribution of chlorophyll *a* at Stn.KNOT

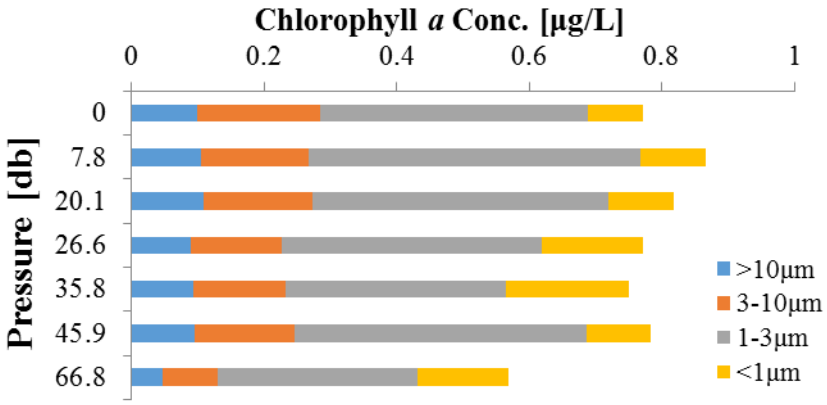


Figure 2.8.1-2 Vertical distribution of size-fractionated chlorophyll *a* (KNOT Cast03)

2.8.2 HPLC measurements of marine phytoplankton pigments

Tetsuichi FUJIKI (JAMSTEC) : Principal Investigator
Keitaro MATSUMOTO (MWJ) : Operation Leader

(1) Objective

The chemotaxonomic assessment of phytoplankton populations present in natural seawater requires taxon-specific algal pigments as good biochemical markers. A high-performance liquid chromatography (HPLC) measurement is an optimum method for separating and quantifying phytoplankton pigments in natural seawater. In this cruise, we measured the marine phytoplankton pigments by HPLC to investigate the marine phytoplankton community structure in the western North Pacific.

(2) Methods, Apparatus and Performance

Seawater samples were collected from eight depths in the euphotic layer at the cast for the primary production. Seawater samples were collected using Niskin bottles, except for the surface water, which was taken by a bucket. Seawater samples (2L) were filtered (<0.02 MPa) through the 47-mm diameter Whatman GF/F filter. The sample filters are stored in a deep-freezer (-80 °C) until onshore HPLC analysis. Phytoplankton pigments are extracted with *N*,*N*-dimethylformamide for at least 24 h at –20 °C in the dark and then will be analyzed with an HPLC modular system (Agilent Technologies) on land.

(3) Data archives

The processed data file of pigments will be submitted to the JAMSTEC Data Management Group (DMG) within a restricted period. Please ask PI for the latest information.

2.8.3 Primary production

Tetsuichi FUJIKI (JAMSTEC) : Principal Investigator
Keitaro MATSUMOTO (MWJ) : Operation Leader

(1) Objective

Quantitative assessment of temporal and spatial variation in carbon uptake in the surface euphotic layer should be an essential part of biogeochemical studies in the western North Pacific. Primary production (PP) was measured as incorporation of inorganic ^{13}C stable isotope tracer at the station of KNOT.

(2) Methods

1) Sampling, incubation bottle and filter

Sampling was conducted at predawn immediately before the incubation experiment. Seawater samples were collected using Teflon-coated and acid-cleaned Niskin bottles, except for the surface water, which was taken by a bucket. Samplings were conducted at eight depths in the euphotic layer in response to the light levels of the incubation containers adjusted with the blue acrylic plate. The light levels of the incubation containers in the bath were shown in Table 2.8.3-1. The light depths relative to the surface had been estimated by the underwater optical sensor on the previous day of the sampling. Seawater samples were placed into acid-cleaned clear polycarbonate bottles in duplicate for PP, and in a single for the dark and the time-zero samples. The time-zero sample was filtered immediately after the addition of ^{13}C solution. Filtration of seawater sample was conducted with pre-combusted glass fiber filters (Whatman GF/F 25 mm) with temperature of 450 degree C for at least 4 hours.

2) Incubation

Each bottle was spiked with sufficient $NaH^{13}CO_3$ just before incubation so that ^{13}C enrichment was about 10% of ambient dissolved inorganic carbon as final concentration of 200 μmol dm^{-3} (Table 2.8.3-2). Incubation was begun at predawn and continued for 24 h. The simulated *in situ* method was conducted in the on-deck bath cooled by running surface seawater or by immersion cooler.

3) Measurement

After 24 hours incubation, samples were filtered through GF/F filter, and the filters were kept in a freezer (-20 degree C). Subsequently, the filters were dried in the oven (45 degree C) for at least 20 hours, and inorganic carbon was removed by acid treatment in HCl vapor bath for 30 minutes. All samples are measured by a mass spectrometer system on land.

(3) Data archives

All data will be submitted to JAMSTEC Data Management Group (DMG).

Table 2.8.3-1 Light levels of the incubation containers

Number	Light Level
#1	100%
#2	43%
#3	14%
#4	6%
#5	3%
#6	0.9%
#7	0.2%

Table 2.8.3-2 Sampling cast table and spike ^{13}C concentration

Incubation type	CTD Station & Cast	$NaH^{13}CO_3$ ($\mu mol\ dm^{-3}$)
simulated *in situ*	KNOT Cast 03	200

2.8.4 FRRF observation

Tetsuichi Fujiki (JAMSTEC)

(1) Objective

During the past decade, the utilization of active fluorescence techniques in biological oceanography brought significant progress in our knowledge of phytoplankton productivity in the oceans. Above all, the fast repetition rate (FRR) fluorometry reduces the primary electron acceptor (Q_a) in photosystem II (PSII) by a series of subsaturating flashlets and can measure a single turnover (ST) fluorescence induction curve in PSII. The PSII parameters, such as the potential photosynthetic activity (F_v/F_m) and the functional absorption cross-section of PSII (σ_{PSII}), derived from the ST fluorescence induction curve can be used to estimate gross primary productivity. In the present study, to gain a better understanding of variability in phytoplankton productivity in the WSG, we measured the PSII parameters and primary productivity using the FRR fluorometry.

(2) Methods

Using the FRR fluorometer (Kimoto Electric Co., Ltd., Japan), the vertical variation in PSII parameters and primary productivity were examined at Sta. KNOT. The FRR fluorometer was moved up and down between surface and 100 m at the rate of 0.2 m s^{-1} using a ship winch. The profiling rate of the observation buoy was set to minimal in order to detect small scale variations (~0.5 m) in measurements. The observation of FRR fluorometer was made at *ca* 10:00 a.m. (LST).

(3) Preliminary results

The profiles of F_m (indicator of chlorophyll *a* biomass), F_v/F_m, and σ_{PSII} at Sta. KNOT measured using the FRR fluorometer were shown in figure 1.

(4) Data archives

All data will be submitted to JAMSTEC DMG and is currently under its control.

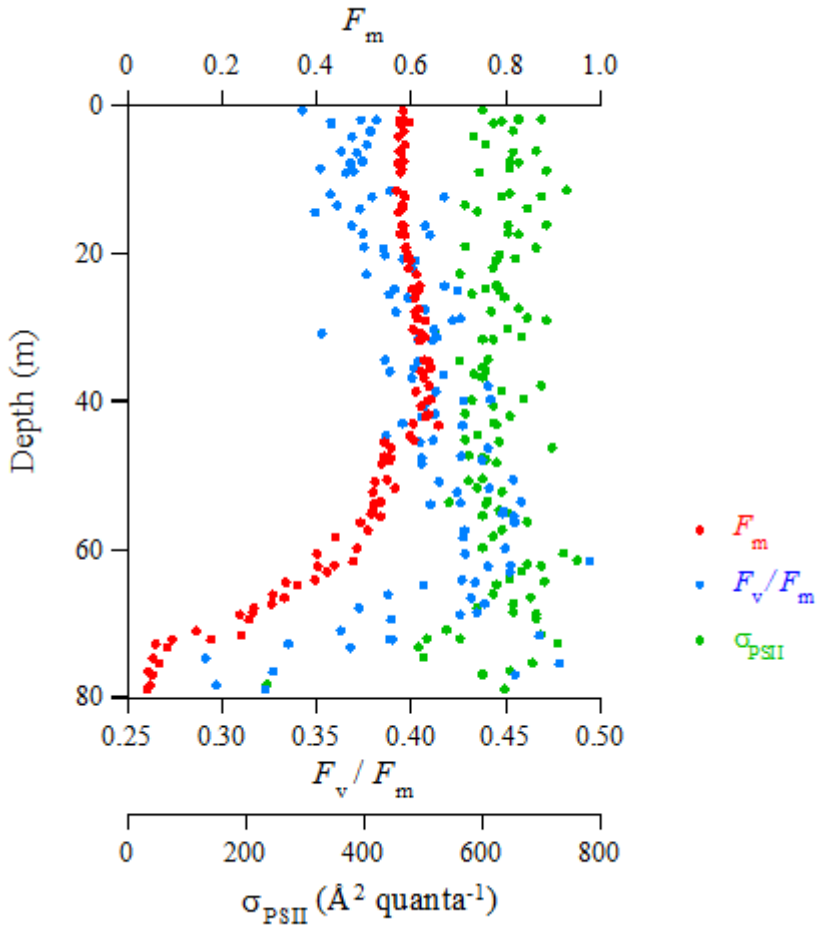


Fig. 1. Vertical profiles of F_m, F_v/F_m, and $_{PSII}$ at Sta. KNOT.

2.9 Zooplankton

Katsunori Kimoto (RCGC, JAMSTEC)
Minoru Kitamura (RCGC, JAMSTEC)
Keisuke Shimizu (Marine Biosciences, JAMSTEC)
Makoto Sugimura (Shin Enoshima Aquarium)

1) Objectives

Owing to the anthropogenic CO_2, concentration of the atmospheric CO_2 is rising. The ocean has already absorbed about 30% of the total anthropogenic CO_2 (approximately 155 GtC) since the industrial revolution (IPCC AR5, 2013). This reduces ocean pH and causes wholescale shift in seawater carbonate chemistry. Ocean acidification also alters biogeochemical processes; one well-known effect is the lowering of calcium carbonate saturation state, which impacts shell-forming marine organisms such as pteropods and planktonic foraminifera. Ocean acidification also affect zooplankton other than the shell-forming organisms, and characteristics of the zooplankton community will be probably changed. So, we are acoustically observing long-term change of zooplankton biomass at K2 by using the mooring system, and need measured values of bulk zooplankton biomass to validate the acoustically estimated biomasses.

In this cruise, for better understanding of biological responses to carbonate saturation status, we aimed for following themes,

(a) to understand vertical distributions of pteropods and planktonic foraminifera communities, and vertical change of their shell densities,
(b) to understand phenotypic and genetic responses against different pCO_2 conditions for pteropods, and their population structure,
(c) to understand influence of water temperature on shell forming of planktonic foraminifera,
(d) trial of long-term rearing for pteropods,
(e) to measure bulk zooplankton biomass at around K2 mooring.

Unfortunately, in this cruise, we could not sufficient pteropod specimens for incubation experiment to understand the phenotypic and genetic responses against different pCO_2 conditions. And we did not collect bulk biomass data of zooplankton at K2 because of the bad weather.

2) Methods

All plankton samplings were conducted not in K2 but in KNOT because of the bad sea state. For the purpose (a), we collected specimens by using the Vertical Multi-depth Plankton Sampler (VMPS-3K, Tsurumi Seiki co., Ltd). VMPS-3K equipped 4 plankton nets (63 μm mesh, NXX25), flow-meter, fluorometer (Wet Lab) and CTD sensors (Sea-bird Electrics), and was hauled vertically at a speed of 0.5 m/sec. Opening/closing of each net is electrically controlled from the lab on the ship, we can

collect vertically stratified sample sets together with the environmental sensing data. Collected samples were immediately fixed by the ethanol (99.5 %) and stored in the refrigerator under 4°C.

For the purposes (b) and (c), a NORPAC net (100 μm mesh, NXX13) with a large cod end was vertically hauled. Living pteropods for the purpose (b, population analysis) were immediately sorted out from NORPAC net samples, and fixed by 99.5 % EtOH. Thereafter, living foraminifera (mainly *Globigerina bulloides*) were picked up for the purpose (c), and they were cultured in filtered seawater at the water temperatures of 4, 7, and 10 ºC and under the dark condition. The water temperatures for the rearing were determined by SST (7–8 ºC) during the samplings. An *Artemia* nauplius was given as a prey to the individual foraminifera once two days. The culture was continued in the land laboratory after the end of the YK16-16 cruise.

Although we could not collect pteropod specimens for the purpose (d), some zooplankton species (krill, copepods, polychaets, and jellyfishes) sorted out from the VMPS and NORPAC net samples were reared in the Kreisel Tank at the water temperature of 4 ºC. Rotifers (*Brachionus plicatilis*) and *Artemia* nauplii were given as prey. The rearing was also continued after the cruise in the Shin Enoshima Aquarium.

3) Onshore study in the future

Pteropod and planktonic foraminiferal shells will be analyzed by the Micro-focus X-ray CT (MXCT) equipped in JAMSTEC HQ in order to elucidate relationships between oceanic carbonate chemistry and individual shell density. For the population analysis of pteropods, DNA extraction from the preserved specimens and amplifying of gene sequences for the COI region of mtDNA will be performed. After the sequence alignments, population genetic analysis will be performed.

(4) Data Archive

All data will be submitted to JAMSTEC Data Management Office (DMO).

2.10 N_2O and CH_4 with stable isotopes

Chisato YOSHIKAWA (JAMSTEC BGC)

(1) Introduction

Nitrous Oxide (N_2O) is recognized as significant anthropogenic greenhouse gas and a stratospheric ozone destroyer. The estimation of global N_2O flux from ocean to the atmosphere is 3.8 $TgNyr^{-1}$ and the estimation varies greatly, from 1.8 to 5.8 $TgNyr^{-1}$ (IPCC, 2013). This is because marine N_2O production processes are poorly understood quantitatively and so previous models had estimated N_2O concentration from oxygen concentration indirectly. Marine N_2O production processes are very complicated; hydroxylamine oxidation during nitrification, nitrite reduction during nitrifier denitrification and nitrite reduction during denitrification produce N_2O and N_2O reduction during denitrification consumes N_2O (Dore et al., 1998; Knowles et al., 1981; Rysgaard et al., 1993; Svensson, 1998; Ueda et al., 1993). N_2O isotopomers (oxygen isotope ratio ($\delta^{18}O$), difference in abundance of $^{14}N^{15}N^{16}O$ and $^{15}N^{14}N^{16}O$ (SP), and average nitrogen isotope ratio ($\delta^{15}N$)) are useful tracers to distinguish these processes and had revealed N_2O production processes in various ocean environments (e.g., Yoshida and Toyoda, 2000). Recently we had newly developed a marine N_2O box model including isotopomers at K2 (Yoshikawa et al., 2015). In this cruise we collected samples for the observed isotope values of N_2O and N_2O related materials, and are planning to expand the model to a one dimensional model by using the isotope data set.

Methane (CH_4) is a terminal product during decomposition of organic matter and acts as a potential greenhouse gas. The vast quantity of methane (about 500-600 Tg) is annually emitted to atmosphere, in which contribution of methane from ocean accounts for 3% (Conrad, 2009). In surface seawater, concentration of methane is oversaturated respect to the atmosphere, where dissolved oxygen concentration is maxima (Scranton et al., 1977). However, the mechanism of the methane production is still ambiguous, which has been termed the ocean methane paradox. As a traditional hypothesis, methane is produced in micro-anaerobic environments such as in guts of zooplankton and sinking particles of organic matter. A recent study suggested bacterial production as byproduct during decomposition of phosphonate (Karl et al., 2008). In this cruise we collected samples for the isotope values of CH_4, and are planning to clarify quantitative distribution of methanogens and their methanogenic pathway, which should provide important information about methane origin in the surface seawater.

(2) Materials and methods

Seawater samples are taken by CTD-CAROUSEL system attached Niskin samplers of 12 L at 12 layers and surface layer taken by plastic bucket at station KNOT.

Samples for N_2O isotopomer analysis was transferred to two of 160 ml glass vials. After an approximately two-fold volume overflow, 100μL of saturated $HgCl_2$ solution were added. The vials were sealed with butyl rubbers and aluminum caps and stored in dark at 4°C until analysis. The $\delta^{15}N$, $\delta^{18}O$ and

SP values and concentrations of N_2O in seawater will be determined by slightly modified version of GC-IRMS (PreCon/HP6890 GC/ MAT 252) at TIT described in detail in Yamagishi et al. (2007).

Samples for NO_3^- and NH_4^+ isotope analysis were collected through GF/F filter. The samples for NO_3^- were removed NO_2^- with sulfamic acid using the method of Granger and Sigman (2009) and preserved at -23°C until chemical analysis. The $\delta^{15}N$ and $\delta^{18}O$ values of NO_3^- will be measured using the "bacterial" method of Sigman et al., (2001) in which N_2O converted from nitrate is analyzed using GasBench/ PreCon/IRMS. The samples for NO_2^- were frozen until analysis. A PTFE pack, which contained a Whatman GF/D filter with 10 μL of 1M H_2SO_4 solution, and 0.3 g of MgO were added to the samples for NH_4^+. The bottles were shaken by hands at least once a day for 5 days. After ammonium was trapped to the acid, the pack was removed and rinsed with MilliQ water, then stored in a glass bottle with silica gels until analysis. The $\delta^{15}N$ values of ammonium will be measured by EA-IRMS.

Samples for $\delta^{13}C$-CH_4 analysis were transferred to 160 mL glass vials from the Niskin sampler without head space. After the vials were sealed with butyl rubber and aluminum caps, 100 μL of saturated $HgCl_2$ solution was added. The water samples were stored until analysis on land. The $\delta^{13}C$ value of Methane will be measured using a method of Tsunogai et al. (1998 and 2000) by IRMS.

Samples for $\delta^{13}C$-DIC analysis were transferred to 30 mL glass vials from the Niskin sampler without head space. After the vials were sealed with butyl rubber and aluminum caps, 20 μL of saturated $HgCl_2$ solution was added. The water samples were stored until analysis on land. The $\delta^{13}C$ value of DIC will be measured by IRMS.

(3) Expected result

In the surface layer, N_2O concentration of water affects the sea-air flux directly (Dore et al., 1998). However the pathway of N_2O production in surface layer is still unresolved. In the surface layer, N_2O is predominantly produced by nitrification, but also by nitrifer-denitrification and denitrification if oxygen concentration is low in the water mass or particles (Maribeb and Laura, 2004). The observed concentrations and isotopomer ratios of N_2O together with those values of substrates for N_2O (NO_3^- and NH_4^+) will reveal the pathway of N_2O production in the surface layer and will expand and improve the marine N_2O isotopomer model.

The $\delta^{13}C$ value of methane reflects carbon source and methaogenic pathway (Whiticar, 1999). If the profile correlate with other chemical profiles including $\delta^{13}C$ value of DIC, concentrations of methane, chlorophyll and dissolved oxygen, it can be a strong evidence of that methanogens are source organisms for methane in the surface seawater. The $\delta^{13}C$ value of methane will support presence of methanogen and provide further constraints on methaongenic pathway.

References

Conrad, R., 2009. The global methane cycle: recent advances in understanding the microbial processes involved. Environ. Microbiol. Rep. 1, 285-292.

Dore, J.E., Popp, B.N., Karl, D.M. and Sansone, F.J.: A large source of atmospheric nitrous oxide from subtropical North Pacific surface water, Nature, 396, 63-66, 1998.

Granger, J., and D. M. Sigman (2009), Removal of nitrite with sulfamic acid for nitrate N and O isotope analysis with the denitrifier method, Rapid Communications in Mass Spectrometry, 23(23), 3753-3762.

IPCC Working group I (2013): Climate change 2013: The physical science basis. IPCC The 5th Assessment report, http://www.ipcc.ch/report/ar5/wg1/.

Karl, M. D., Beversdorf, L., Björkman, M. K., Church, J. M., Martinez, A. and DeLong, F. E., 2008. Aerobic production of methane in the sea. *Nature Geoscience* **1**, 473-478.

Maribeb, C.-G. and Laura, F.: N_2O cycling at the core of the oxygen minimum zone off northern Chile, Marine Ecology Progress Series, 280, 1-11, 2004.

Rysgaard, S., Risgaard-Petersen, N., Nielsen, L.P. and Revsbech, N.P.: Nitrification and denitrification in lake and estuarine sediments measured by the ^{15}N dilution technique and isotope pairing, Applied and Environmental Microbiology, 59, 2093-2098, 1993.

Scranton, I. M. and Brewer, G. P., 1977. Occurrence of Methane in near-Surface Waters of Western Subtropical North-Atlantic. Deep-Sea Research 24, 127-138.

Sigman, D. M., K. L. Casciotti, M. Andreani, C. Barford, M. Galanter, and J. K. Boehlke: A bacterial method for the nitrogen isotopic analysis of nitrate in seawater and freshwater, Anal. Chem., 73, 4145-4153, 2001.

Svensson, J.M.: Emission of N_2O, nitrification and denitrification in a eutrophic lake sediment bioturbated by Chironomus plumosus, Aquatic Microbial Ecology, 14, 289-299, 1998.

Tsunogai, U., Ishibashi, J., Wakita, H. and Gamo, T., 1998. Methane-rich plumes in the Suruga Trough (Japan) and their carbon isotopic characterization. *Earth. Planet. Sci. Lett.* **160**, 97-105.

Tsunogai, U., Yoshida, N., Ishibashi, J. and Gamo, T., 2000. Carbon isotopic distribution of methane in deep-sea hydrothermal plume, Myojin Knoll Caldera, Izu-Bonin arc: Implications for microbial methane oxidation in the oceans and applications to heat flux estimation. *Geochim. Cosmochim. Acta* **64**, 2439-2452.

Ueda, S., Ogura, N. and Yoshinari, T.: Accumulation of nitrous oxide in aerobic ground water, Water Research, 27, 1787-1792, 1993 www.cia.gov/cia/publications/factbook/geos/zn.html

Yamagishi, H., Westley, M.B., Popp, B.N., Toyoda, S., Yoshida, N., Watanabe, S., Koba, K., Yamanaka, Y., 2007. Role of nitrification and denitrification on the nitrous oxide cycle in the eastern tropical North Pacific and Gulf of California. J. Geophys. Res. Biogeosciences 112, n/a–n/a. doi:10.1029/2006JG000227

Yoshida, N. and Toyoda, S.: Constraining the atmospheric N_2O budget from intramolecular site preference in N_2O isotopomers, Nature, 405, 330-334, 2000.

Yoshikawa, C., H. Abe, M.N. Aita, F. Breider, K. Kuzunuki, N.O. Ogawa, H. Suga, N. Ohkouchi, S. O. Danielache, M. Wakita, M.C. Honda, S. Toyoda, N. Yoshida, 2016. Insights into the production

processes of nitrous oxide in the western north Pacific by using a marine ecosystem isotopomer model, Journal of Oceanography, DOI:10.1007/s10872-015-0308-2.

Whiticar M.J., 1999. Carbon and hydrogen isotope systematics of bacterial formation and oxidation of methane. *Chem. Geol.* **161**, 291-314.

2.11 Increase of N_2O production rates as a consequence of ocean acidification

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(1) Introduction

Oceanic N_2O production is dominated by nitrification and represent ~30% of the total source of atmospheric N_2O (Freing et al., 2012; Codispoti, 2010). The uptake of anthropogenic CO_2 by oceans might lead to a reduction of seawater pH ranging from 0.2 to 0.5 units below the preindustrial level by the year 2100 (Caldeira and Wickett, 2003). Surface ocean pH is already about 0.1 unit lower than pre-industrial values. Therefore, if anthropogenic CO_2 emissions continue at the same rate, the rate of change of ocean pH would be faster than changes detected in geological records (Kump et al., 2009; Pearson and Palmer, 2000). Marine N_2O production may change substantially as a result of oceans acidification. As the NH_3/NH_4^+ equilibrium is pH-sensitive and ammonia-oxidizing bacteria (AOB) and archaea (AOA) use preferentially NH_3 rather than NH_4^+, nitrification reaction might be partly inhibited under reduced pH. Recently Beman et al. (2011) have shown that ocean acidification could reduce ammonia oxidation rates in open ocean by 3 to 44% within the next few decades. Based on nitrification rate measurements, Beman et al. (2011) have estimated that the decrease of the nitrification induced by ocean acidification would lead to a reduction of N_2O production ranging from ~0.06 to 0.83 Tg N yr^{-1}. Such reduction of the oceanic N_2O emission might be comparable to all current N_2O production from industrial processes and fossil fuel burning (IPCC, 2007). Despite of the clear influence of the pH on ammonia oxidation rate, so far no study has investigated directly the effect of pH change on N_2O production rates in the oceans. It is still unknown how the decline of oceans pH induced by rising of atmospheric CO_2 concentration will affect oceanic N_2O emission rates.

(2) Materials and methods

Seawater samples are taken by CTD-CAROUSEL system attached Niskin samplers of 12 L at 100 m and 150 m depths at station KNOT. Seawater samples were sub-sampled into two 200 mL glass vials. The samples were acidified with different amount of 1 M HCl solution. Then $^{15}NH_4Cl$ solution was added in seawater samples using a microsyringe, so that the final concentrations were 50 or 250 nmol N L^{-1}. The samples were incubated in the dark at a temperature close to that at the sampling depth during 12h and 24h. After incubation, 20 ml of each sample was removed for ammonium oxidation rate analysis and pH measurements and this aliquot of 20 ml was compensated by adding degased MilliQ water. Then the samples for N_2O production rates were sterilized with 100 μL of saturated $HgCl_2$ solutions and stored in the dark at 4°C until analysis. The samples for ammonia oxidation rates were filtered with DISMIC® filter (0.45 μm) and stored at -20°C until isotopic analysis. The pH of seawater samples measured using a pH electrode.

The $\delta^{15}N$, $\delta^{18}O$ and SP values and concentrations of N_2O in seawater will be determined by

slightly modified version of GC-IRMS (PreCon/HP6890 GC/ MAT 252) at Tokyo Tech. described in detail in Yamagishi et al. (2007). The $\delta^{15}N$ and $\delta^{18}O$ values of NO_3^- will be measured using the "bacterial" method of Sigman et al., (2001) in which N_2O converted from nitrate is analyzed using GasBench/ PreCon/IRMS at JAMSTEC.

(3) Expected result

In the subarctic western North Pacific, the ammonia oxidation rates and N_2O production rates were 33 and 0.2 nmolL-1day^{-1} at 100 m depth in July, 2013 (Yoshikawa et al., 2016). Those rates are significantly high relative to those rates in the subtropical western North Pacific. Nitrification is considered to increase activity from autumn to winter. The ammonia oxidation rates and N_2O production rates in this series of incubation experiments must be detectable and we can assess the impact of ocean acidification on N_2O production rates.

References

Freing, A., Wallace, D. W. R. & Bange, H. W. Global oceanic production of nitrous oxide. Philos. Trans. R. Soc. B Biol. Sci. 367, 1245–1255 (2012)

Codispoti, L. A. Interesting Times for Marine N_2O. Science 327, 1339–1340 (2010).

Caldeira, K. & Wickett, M. E. Oceanography: Anthropogenic carbon and ocean pH. Nature 425, 365–365 (2003).

Kump, L. R., Bralower, T. J. & Ridgwell, A. Ocean Acidification in Deep Time. Oceanography v.22(4), (2009).

Pearson, P. N. & Palmer, M. R. Atmospheric carbon dioxide concentrations over the past 60 million years. Nature 406, 695–699 (2000)

Beman, J. M. et al. Global declines in oceanic nitrification rates as a consequence of ocean acidification. Proc. Natl. Acad. Sci. 108, 208–213 (2011).

Yamagishi, H., Westley, M.B., Popp, B.N., Toyoda, S., Yoshida, N., Watanabe, S., Koba, K., Yamanaka, Y., 2007. Role of nitrification and denitrification on the nitrous oxide cycle in the eastern tropical North Pacific and Gulf of California. J. Geophys. Res. Biogeosciences 112, n/a–n/a. doi:10.1029/2006JG000227

Sigman, D. M., K. L. Casciotti, M. Andreani, C. Barford, M. Galanter, and J. K. Boehlke: A bacterial method for the nitrogen isotopic analysis of nitrate in seawater and freshwater, Anal. Chem., 73, 4145-4153, 2001.

Yoshikawa, C., H. Abe, M.N. Aita, F. Breider, K. Kuzunuki, N.O. Ogawa, H. Suga, N. Ohkouchi, S. O. Danielache, M. Wakita, M.C. Honda, S. Toyoda, N. Yoshida, 2016. Insights into the production processes of nitrous oxide in the western north Pacific by using a marine ecosystem isotopomer model, Journal of Oceanography, DOI:10.1007/s10872-015-0308-2.

3. Tsunami early warnings by the seafloor electromagnetic observations

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Purpose and backgrounds

The objective of the second proposal is to recover SFEMS5 (Sea-Floor Electro-Magnetic Station 5) that was deployed at North-West Pacific site (NWP) in May, 2013, by the research cruise of KR13-09. SFEMS5 was equipped with a differential pressure gauge (DPG), for detection of tsunami-generated electromagnetic (EM) and pressure variations simultaneously at the seafloor. It was recently revealed that the seafloor EM variations are useful to investigate kinetic properties of tsunamis. For instance, in oceans deeper than 5000m, the vertical component of the magnetic field is in phase with tsunami height and a measure of tsunami wave height, while variations of the horizontal components enable measurements of tsunami propagation directions. Combination of the long-term seafloor EM and seafloor pressure data will contribute to more detailed understandings of the relation between tsunamis and EM fields and to the developments of tsunami early warning system using EM variations.

Observations

On November 14th, 2016, we succeeded in retrieving SFEMS5 at 09:01 am (JST), after acoustic release at 04:37 am (JST). It took 4 hours for SFEMS5 to ascend from the seafloor up to the sea surface. Any glass sphere flooding was not found, which is a remarkable success considering such a long observation duration of over 3.5 years. When SFEMS5 was retrieved, the battery already ran out. However, the internal clock time of both the interface and the ocean bottom electromagnetometer (OBEM) was obtained by using an external power source. It was revealed that the internal clock had gained ~3 min since deployment3.5 years ago.

Methods and instruments

SFEMS usually consists of the 5 main 17'' glass spheres of interface, battery, gyro, OBEM, and Overhauser magnetometer. Additionally, SFEMS5 was equipped with DPG in order to detect tsunami height directly. An ROV homer (an acoustic transponder) was arranged for SFEMS5 in KR13-09 to guarantee quick spotting by ROV at the seafloor. All the instruments attached to SFEMS5 were recovered safely by acoustic release.

Research results

We obtained three components of geomagnetic data and two horizontal components of geoelectric data for 2.8 years with a sampling interval of one minute. Unfortunately, the magnetic total intensity record by Overhauser magnetometer was limited to the first 80 days. From all the four-time gyro measurements, we

inferred that the north axis of the instruments pointed in the direction of N32.27^{o}W at the seafloor. Although it is uncertain whether the obtained data include tsunami signals at the present, such long-term continuous EM time series data at open ocean will no doubt provide us with precious opportunity to investigate not only tsunami-related EM variations but also Earth's internal structure, the secular variation of the geomagnetic main field, and couplings of EM fields with other oceanic flows than tsunamis.

4. Ocean depth sounding just after the off Fukushima Earthquake on Nov. 22nd, 2016

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Purpose and backgrounds

During the present cruise, a tsunamigenic earthquake (M7.4) occurred off Fukushima prefecture at 05:59:58 JST, November 22nd, 2016. According to Japan Meteorological Agency (JMA), tsunami reached eastern coastlines of the Northeast Japan. For example, 1.4 m tsunami height was reported at Sendai Port. Hi-net AQUA system of National Research Institute for Earth Science and Disaster Prevention (NIED) rapidly inferred the location of hypocenter as (141.5°E, 37.3°N, -11km) and the strike of the fault plane as N51.2°E. When the tsunami event occurred, the observations planned for YK16-16 were already finished so that Yokosuka was heading back to the Yokosuka port. As there was one more available day, we decided to make an acoustic bathymetry survey around the focal area to investigate the seafloor crustal deformation due to the tsunamigenic earthquake.

Observations

We conducted an acoustic survey of ocean depth on November 23rd, focusing on an area of 3 mile × 3mile centering the epicenter estimated by NIED. Although there were several reports of focal mechanisms/epicenters that differed in position by a maximum of 0.2 degree, we chose the first position estimated by NIED as the center of our survey area because it was close to the averaged position for all the reported epicenters.

Methods and instruments

For the ocean depth survey, we used MultiBeam Echo Sounder (MBES) equipped with R/V Yokosuka. The area of 3 mile × 3 mile was surveyed by 13 north-south profiles with a length of 3 miles each and with a east-west interval of 400 m. The survey width by MBES was limited to 400 m due to the shallow ocean depths from 150 to 200 m around the focal area.

Research results

We succeeded in obtaining high-resolution 3 mile × 3 mile bathymetry data in the focal area one day after the off Fukushima Earthquake on November 22, 2016. The horizontal spatial resolution is ~10 m while the resolution of the ocean depth is ~1m, respectively. From the data, we clearly recognized an upheaval structure with a north-south extent of 2 km along a latitude line of 37.28° against the background plain bathymetry deepening eastward to the Japan Trench. The obtained ocean depth dataset can be compared to the past/future datasets to estimate the seafloor crustal deformation due to the tsunamigenic earthquake.