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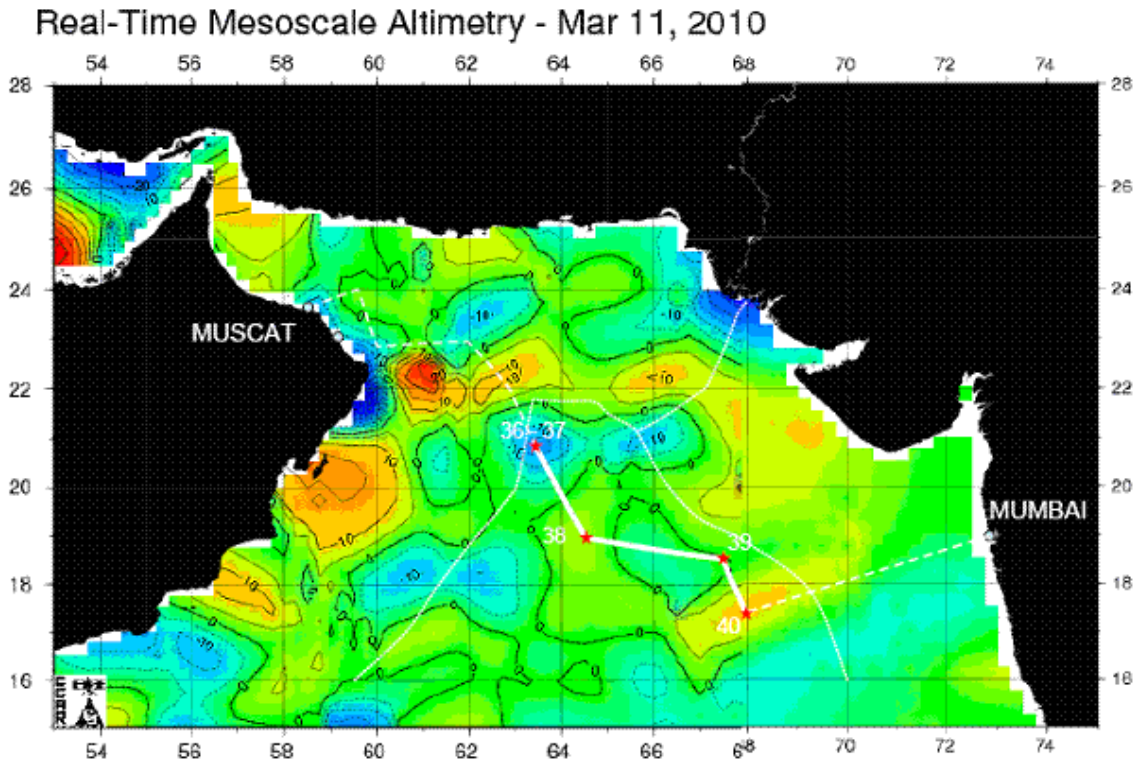
CHIEF SCIENTIST'S SUMMARY REPORT

CRUISE OBJECTIVES

The objectives were to sample the Arabian Sea in the North Indian Ocean from Muscat to Mumbai in order to collect and describe planktonic organisms ranging from viruses to macro-zooplankton. In the same time, a description of the water masses encountered during the cruise was also undertaken in order to replace the living organisms in their physico-chemical environment. This was absolutely necessary to interpret the data in terms of functional diversity. A special attention was also given to the Oxygen Minimum Zone which is a recognized particularity of this part of the North Indian Ocean, as it constitutes the major reservoir of anoxic water of the global ocean. For this reason, this zone was extensively sampled.

MAJOR ACHIEVEMENTS & FINDINGS

AS Tara was not authorized to sample in the Oman and Indian seas, and because of the pirate pressure coming from Somalia, the investigation zone was restricted to a relatively small central triangle in the International waters of the Arabian Sea : 18.37°N 61.54°E – 21.35°N 63.22°E – 18.48°N 67.31°E, in which four main 2 days stations were sampled during this leg. The first station day was a standard Core Station, the second day was essentially devoted to repetitive Niskin sampling with the rosette to provide large volumes of deep sea water taken in the Oxygen Minimum Zone for virus and bacteria and protist analyses. The relatively heavy schedule was possible only because of the excellent meteorological conditions experienced during the cruise.



Station locations were chosen using the sea surface height anomalies maps kindly provided by Dr. Serguey Piontkovski met onboard of Tara during the Muscat stopover. It was thus possible to sample

in contrasted trophic zones associated to different cold, warm, and in between mesoscale eddies, in which it was expected to find (and actually observed) different organisms and communities, associated to different ecosystemic functions (production vs regeneration). We also benefited from the Gilles Reverdin (LOCEAN) Argos buoy which was onboard. The floating anchor and the buoy, deployed at the beginning of each station, allowed the regular repositioning of Tara in the same water mass during the 48 hours sampling and profile acquisitions spent at each station. The other novelty during this leg was the successful deployment of the Multinet, after only 2 unsuccessful trials.

MAJOR CONCERNS & ACTIONS TO TAKE

This leg could be depicted as a geopolitics-scientific operation. It can be said without exaggeration that the lack of visibility on the possibility to sample in the Oman and Indian waters at the starting and the ending of the cruise respectively did not help to optimize the time-ship for two reasons. First because we missed very highly productive coastal and interesting sites in the Oman straight and Indian continental shelves; second because it was very difficult to optimize the sampling schedule, which normally should be more or less established at the beginning of the leg.

So for the coming cruises, I really believe that all the authorizations (visa, sampling, and place in the port) should be imperatively obtained before the departure of the ship.

From an operating point on view, the oceanographic part of the mission would take a great advantage to be regularly provided with MODIS maps of sea-surface chlorophyll, and altimetry anomalies. In the same way, the permanent installation on board of a floating anchor (not necessary coupled with and Argos buoy) would greatly improve the quality of the sampling, notably in dynamics zone with high spatial mesoscale variability.

PARTICIPANTS

	ROLE	**NAME, Surname, Affiliation**
1	CREW- Captain	BOURMAUD Hervé, Captain
2	CREW- 1st Officer	MENARD Mathilde, Chief mate
3	CREW-	BRACQ Guillaume, Chief engineer
4	CREW-	CRON Daniel, 2nd engineer
5	CREW-	GIESE Alain, Deck officer
6	CREW- Cook	GIRARDOT Julien, cook
7	CREW- Media	BASTION Jérome, journalist
8	MEDIA-	TEIGNE Jérome, cameraman
9	MEDIA-	BROUTIN Olivier,
10	SCIENCE- Chief Scientist	SCIANDRA Antoine STEPHANE Pesant
11	SCIENCE- Oceanography Engineer	PICHERAL Marc
12	SCIENCE- Optical Engineer	GILETTE Jennifer
13	SCIENCE- Biology Engineer	DIMIER Céline
14	SCIENCE- Biology	ARSLAN Defne
15	SCIENCE- Backup	

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GENERAL DIVISION OF WORK

RESPONSIBILITIES	**NAME**
LOGISTICS – Planning, quality assurance, data & metadata archives	Chief Sci. SCIANDRA and PESANT
LOGISTICS – Consumables and samples storage & inventories	Biol. Eng. DIMIER
BGC – Nutrients	Chief Sci. SCIANDRA
BGC – Carbonates	Chief Sci. SCIANDRA
BGC – Hg	Chief Sci. *NOT SAMPLED*
BGC – OM Lugol/Formol	Chief Sci. SCIANDRA
BGC – Cultures	Chief Sci. SCIANDRA
BGC – HPLC	Chief Sci. PESANT
META – Metagenomic	Opt. Eng. GILETTE
META – Taxo Genetic	Opt. Eng. GILETTE
META – Taxo Morphology	Opt. Eng. GILETTE
BACT – GIRUS – VIRUS	ARSLAN
PROT – dDNA	DIMIER
IMAG – FlowCam	Opt. Eng. GILETTE
IMAG – Macroscopy	Opt. Eng. GILETTE
IMAG – SeaFlow	Opt. Eng. GILETTE
IMAG – SPIM	Opt. Eng. *NOT USED*
OCEANO – Rosette	Oceano. Eng. PICHERAL
OCEANO – Nets	Oceano. Eng. PICHERAL
OCEANO – Pump	Oceano. Eng. PICHERAL
OCEANO – TSRB	Oceano. Eng. PICHERAL
OCEANO – ARGO floats	Oceano. Eng. PICHERAL
OCEANO - TSG	Oceano. Eng. PICHERAL
OCEANO – FRRF	Oceano. Eng. PICHERAL
OCEANO – ACS	Oceano. Eng. *NOT USED*

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CALENDAR OF ACTIVITIES

DATE (MMDD)	ACTIVITY	COMMENT
9 March	Departure from Muscat	
10 March	Route and sea surface Bucket sampling	Preparation planning of the next station
11 March	Route and sea surface Bucket sampling	
12 March	Core Station	ST36, Argos Buoy deployment
13 March	Deep Sampling Rosette Station	ST37, Multinet trials, Argos Buoy recovery
14 March	Route	A lot of macroplankton visible from the ship deck, especially large Salpa chains, and jellyfish
15 March	Core Station	ST38, Argos Buoy Deploy. Multinet trials
16 March	Deep Sampling Rosette Station	ST38, Argos Buoy Recovery
17 March	Route, Continuous Seawater pump fixed (no data)	Reparation of water closet, fuel leak in the carre,
18 March	Core Station	ST39, ARGOS deploy, cable of the main winch broken twice in the morning, no loosen material. ARGOS recover.
19 March	Multinet tests (OK)	ST39
20 March	Deep Sampling Rosette Station	ST39. No repositioning of Tara. Multinet OK
21 March	Route	
22 March	Water column sampling with rosettte	ST40 ARGOS deployement
23 March	Route	Boat cleaning and relaxation
24 March	Route	
25 March	Mumbai arrival	

GENERAL ASSESSMENT, SPECIFIC CONCERNS & ACTIONS TO TAKE

NAME: Sciandra Antoine

(if anonymous, the comments will be taken into consideration in the chief scientist's report)

LIFE ONBOARD

Chief of Mission (Antoine Sciandra). The life onboard was, in my opinion, very pleasant. The relations with all the crew members were excellent, as all of them showed a spirit of strong cooperation. The scientific team was greatly assisted in its quotidian job, both for the deck operations but also for fixing different things. Humour and jokes were our quotidian. The food was excellent, despite the lack of wine. If I omit to consider the toilets reparation and the fuel leak in the carre which occurred simultaneously and delivered a curious mixing of odors, the quality of life on board was very good.

Optical engineer (Gennifer Gilette). Overall I was quite impressed by the accommodations onboard Tara, although it did get quite warm onboard. This is a small vessel, so it is inevitable that there will be problems, and I think we did experience a number of issues during our leg. In particular the AC was not working in the dry lab for 2 days, which resulted in 35-36deg temperatures. This of course is not good for the equipment. However, with each issue the crew stepped to the challenge and we were able to overcome every problem. The AC was repaired by establishing a new seawater line for the AC unit. Overall, it was a pleasure working with the captain and the crew and I was impressed by their hard work and dedication.

Biologist engineer (Celine Dimier). Life onboard was OK. There were several technical problems on the boat (toilets blocked, fuel leak, etc…) that perturbed a little bit the life during 2 days. Nevertheless, the crew, as well as some other people, did their best to quickly solve the problems.

During the leg, tension appeared particularly because of the interdiction to sample neither in the Oman waters nor in the indian waters. In addition, tension increased since we did not know if we could get off in Mumbaï, or if we had to go directly to Male, which meant 12 days more at sea.

Oceanography engineer (Marc Picheral). Difficult due to warm climate. The ship needs to be ventilated. The temperature in the library remained between 30°C and 36°C during all leg.

COMMUNICATION (ONBOARD and ASHORE)

Chief of mission (Antoine Sciandra) Mailing communication was the weak point of this cruise; notably, it was not possible to exchange emails between the different mailing boxes of Tara (captain, engineer, science), neither it was possible to receive and send files bigger than 200 Ko. This was particularly prejudicial to this leg as the sampling strategy was based on the examination of different weekly actualized maps received from different labs (Chlorophyll, Altimetry, Currents).

Besides this aspect, the captain organized different useful informative meetings, although it seems that he was not himself regularly informed on the problems that Tara encountered during the cruise (sampling and stopover authorizations). The sate of ignorance in which all the people on Tara were 3 days before the cruise ending is the sign of inefficient communication.

OIptical engineer (Gennifer Gillette). Scientific Communication: For me, this part of the expedition has been a bit disappointing. There has been minimal scientific discussion during the leg. It seems that this lack of communication was felt throughout the group and it was clearly evident on occasions where station numbers weren't clear, the timing of different deployments wasn't known, or the reasoning behind the station wasn't understood. While in my opinion, this isn't the most productive way to maintain the hardworking morale of a group, I also admit that I am not experienced enough to know the expected level of discussion onboard cruises. Despite this disappointment, it was a pleasure to be part of such a hardworking and motivated scientific group. We had a number of issues during our time on board, but this didn't affect the scientific productivity in the slightest. This can be attributed to the hard work of both the scientists and the crew.

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The International Expedition: Probably my biggest disappointment regarding my time onboard Tara was my inability to communicate with my colleagues. I blame myself for this. If I had been a little more aware, I would have spent my last few months of preparation working on my conversational French. I assumed that daily conversations would be in French, however, I was not prepared to have all the instructions on deck and the science to be in French. I think it is wise to make non-French speakers aware of this fact before they arrive. I took French in school and while I can read and speak slightly, I was not prepared for the level of French required to communicate effectively. I apologize that people had to regularly reiterate information to me in English.

Biologist engineer (Celine Dimier). Communication was sometimes hard, between the scientists themselves, and between crew and scientists. The aim of each sampled station was not always clear in my mind.

Oceanography engineer (Marc Picheral). Email worked fine after initial restart of the server by the captain

SECURITY

Chief of Mission (Antoine Sciandra). Nothing to say, security briefing was made by the captain and second captain, who regularly recall the crew and scientists to use the life jacket during deck operations or zodiac utilization.

Biologist engineer (Celine Dimier). Security rules have been clearly explained when I arrived onboard. They were quite well respected by everyone (lifejacket, secure shoes, etc…)

SAMPLING STRATEGY

Chief of Mission (Antoine Sciandra). The sampling strategy has been adapted to the state of uncertainty in which the chiefs of mission were, notably concerning the areas authorized to be sampled and the pirate presence in the south. So, no initial global sampling strategy could be adopted, as it is generally the case before any oceanographic cruise departure, and we have had to decide the route and the next position at the end of each station.

Because the authorized working area was finally relatively small relatively to the cruise duration, it was possible to choose stations not necessarily aligned along the shortest route between Muscat and Mumbai. The stations were chosen according to the presence of warm and cold mesoscale eddies which presumably support different biological activities, and consequently different communities of planktonic organisms. We were helped in the localization of these structures, by the MODIS maps kindly provided by Serguey Piontkovski, which were much more precise than the Mercator sea surface predictions. By this way, we increased the probability to explore and sample a wider range of biodiversity. The sampling strategy had also to take into account the presence of a very deep and widespread anoxic layer (~150-1000m), a major characteristic of the North Indian Ocean.

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Each station excepted the last one spent 2 days, the first one being a “classical core station”, and the second one being devoted to sample large volumes of deep anoxic seawater. An Argos buoy was deployed and recovered at each station, which allowed Tara to follow the same water body during the two days station. This was very important to be confident in the time variations of physico-chemical properties observed over the 2 day sampling, and to guarantee homogeneous repeat sampling.

The multinet depths were chosen to selectively sample above, in and below the anoxic layer, taking into account also the under water profiler profiles to focus on intermediate layers characterized by high particle content.

The counterpart of this strategy was, noticeably for Marc, Defne and Cecile, a very dense schedule, with 2 consecutive working days, followed by one day during which the next station was prepared. Fortunately, the contribution of the crew and the nice weather helped us greatly.

Biologist engineer (Celine Dimier). Sampling strategy was hard physically at the beginning, with 3 long stations and 2 long stations with 1 depth sampled in 7 days. After, the timing was lighter, with one station per day, followed by a day off. Nevertheless, I think we did a good job, with very different areas sampled.

Oceanography engineer (Marc Picheral). Except a brief initial meeting, everything was decided by the Chief Scientist and Stephane Pesant at the last minute with very limited consideration to other scientists ending with 20h working days. The recovery day between two stations is a busy day for deck engineer processing data and fixing technical problems that always occurs when managing so many instruments at sea.

STATION PREPARATION

Chief of Mission (Antoine Sciandra). Station preparation was mostly under the supervision of Stéphane Pesant, according to a well established protocol known by him; the system of work sheet and bar-coding is very efficient when people are familiarized with it, but not evident *a priori.* This let me think that at least one day of exchange during the stopover should be necessary between the ancient and the new chief of mission.

Oceanography engineer (Marc Picheral). Planning was delivered by the leading team at the last moment and not discussed. I had to be ready for everything all time.

ACTIVITIES ON DECK (e.g. instruments, protocols, timing)

Chief of Mission (Antoine Sciandra). Activities on deck were very intense as all the devices (rosette, plankton nets, pumping, TSRB, Secchi disk…) present on Tara were used repeatedly. The very clement

state of the sea allowed us to respect the scheduled timing of core stations which started at 7:30 and ended at 24:00. The novelty was the deployment of an Argos buoy attached to floating anchor. The GPS position was received and retransmitted to Tara via the LOCEAN. We also deploy the Multinet. After 2 unsuccessful trials and a modification by Marc (lest added), it was possible to use it in routine (until 1200m depth).

There is no doubt for me that the height of the A-frame is just sufficient to deploy the rosette and the Multinet in safety conditions, and that 20 cm more would not be a luxury. During the deployment of plankton net and Argos buoy, the cable of the main winch was broken twice for a reason that was not elucidated yet, fortunately without dramatic consequences due to excellent reactivity of Marc. But if the rosette, which is much heavier, was deployed at this time, the rest of the mission would have been compromised.

Biologist engineer (Celine Dimier). Timing of sampling (i.e. net sampling) was done in agreement with the deck engineer and me. Chief scientist always asked me if I was ready before net sampling with the 20 and 180 um nets.

Two nets of the GPSS had to be repaired because of small holes. We put some glue to fix them.

I personally think that the cooler used for sampling storage on the deck is not sufficient to keep samples in a cool environment. With the heat, ice packs thaw rapidly. We don't have enough ice packs and during the station, we don't have time to go to the freezer to bring each sample.

Oceanography engineer (Marc Picheral). The working rhythm of two deck working days and a single recovery day between long stations is too high and not adapted to the ship working capability. The winch crew and a second crew helped the oceano engineer to deploy the gears except the last day. The last working day was thus dangerous and tiring on deck maneuvering the gears. A TRUE resting time MUST be included in the long station sampling day as all operations cannot rely on a 20h working day for the engineer. The danger increases a lot with tiring.

The Multinet is now working fine. As a rather heavy instrument, it requires more crew on deck and must be deployed early after the sunset when people can help.

ACTIVITIES IN THE WETLAB (e.g. instruments, protocols, timing)

Defne Darlan.

<u>Protocole</u>

Some mistakes between protocoles and logsheet :

Tanit: Fish samples are shared between Virus and Tanit (3+3), and because we put the 3 filters in one petrislide, just one box is sufficient in the logsheet. The FISH line is missing in Virus logsheet.

I had the time to filter 2 replicates from DCM for DNA/RNA Tanit because the Girus filtration units are free during this time. This should be discussed if the filters are for TANIT or GIRUS.

Girus: No DNA for DCM, just culture. For the last stations, I filtered the 100L just in one GFA and Express Plus. However, when the filtration became slow, I changed first the the Express plus.

Errors in the Working box for TANIT/VIRUS/GIRUS:

See Excel attached file (corrections made for core station and new for deep station)

Main observations

Rupture of GFA : Avoid air!! Filter broke quickly in case of air in the tube => Check the tube in the tank. In case of air in the inflow tube, open the valve before it comes to the filter.

Girus Pump Head : I changed the Masterflex tube because it seemed to be damaged and making air bubbles. And it seems to be less efficient than the Tanit pump head.

Iron Chloride : I did it in the Girus first filtration unit and with an inflow tube just for Iron. Avoid contamination, the filtration unit has to be rinsed well with MilliQ water after use. It took around 40 minute to filter 20l.

About the MOZ (water from rosette), when the water is shared between protist and Tanit/Virus.Girus, it's better to begin with Girus in Girus filtration unit to have more time for Iron at the end.

Blank test: 100L of filtered seawater through GFA/Express plus, 20L of this filtrate for Iron chlorride, and 20l through 0.1µm for Girus. It should be done between two stations.

I propose this succession of events:

First, begin with RNA filtration in TANIT pump head and filtration unit. Just after launch the DNA filtration for Girus in Girus filtration unit. When RNA is finished, begin with DNA. Collect the filtrate for Virus and Girus. Fix FISH sample. Process the FCM-Glu & culture sample. Collect water for Virus : qPCR, Culture, TEM and SYBR.

If you have time begin with the 20L of Girus or do it between Surface and DCM.

Then continue with replicate 2 and at the end filter the iron chloride.

Biologist engineer (Celine Dimier). The wetlab is not enough ventilated. Since we use a lot of formol, and that formol is stored in the cupboard, it would be good to find a way to deal with this problem. Sharing the wet lab with the girus person, as well as the chief scientist, was not a problem.

We still have a problem with the protist peristaltic pump since a lot of black particles are found on the 0.8 um filter. We did a comparison with the peristaltic pump used for girus/virus/bacteria, where no black particles were found.

ACTIVITIES IN THE DRYLAB (e.g. instruments, protocols, timing)

Optical Engineer (Jennifer Gillette – Email: gillettj@mail.nih.gov). FlowCam: The FlowCam works great. I had only one error during the entire leg. However, the back up HD is not working reliably. The HD is often not recognized by the FlowCam computer. I tried replace all of the cables with hopes

of identifying the problem, but was unable to find the issue. I have been able to back up all data except for Stn040. In addition the sample funnel on the system has a leak that seems to be getting progressively worse. I went to the replacement parts to replace the funnel, but that funnel is also broken. The leak seems to occur at the funnel head. This should be replaced or it the pieces could be glued together. Finally, the flow cell is starting to look very dirty with streaks and they seem to be getting worse with use. If the flow cell cannot be cleaned well, I would recommend replacing the flowcell before imaging again.

Seaflow: I was unaware that I was responsible for this instrument prior to my arrival. As such, this was an initial point of frustration. The connection link with Washington State was critical because it was only through this communication that I was able to get the system running without proper training. When the system was finally running it worked great, however, the laser and flow alignment are not stable, which resulted in poor data acquisition. This needs to be addressed because the optical engineer cannot devote the time required on a daily basis to maintain the necessary alignment. Currently, the green laser on the system is no longer working. It will not turn on even after a manual switch. It is still unclear as to the cause of the laser failure, but that is still being addressed. Now that the seawater line has been changed for the AC in the dry lab, the water pressure entering the Seaflow is too high (approximately 8 psi and it shouldn't be above 5psi). This will also need to be addressed for future use of the instrument.

SPIM: The SPIM is a mess. Upon my arrival, it was covered with dirt and dust. Some kind of cover is clearly required to protect the system. There is also a power outlet problem in the dry lab. Currently, there are not enough outlets to have the SPIM running with all of the other equipment. 3 outlets are required for the SPIM, yet only 1 outlet remains available. This will need to be addressed for future use. Also, the USB port in the back of the SPIM computer that connected the system to the monitor and keyboard is broken. In order to use the monitor and keyboard, we had to connect the system through the USB port on the front of the computer.

Macroscope: The scope works well, but nothing has been standardized and the scale bar really needs to be set on the machine. It would be nice to include a scale bar with every image acquired. I had difficulty establishing a nice dark, black background on the system. A nice optimized cloth background should be included in with the imaging equipment. I still have not been able to find the Canon 5D camera in the dry lab or in the benches in the small carre.

Zooplankton: All of the net collections went well, however a clear protocol should be established for nets with more than 250ml of biomass. We tried 2 different protocols: 1) using multiple bottles to keep everything from each net and 2) we also tried cleaning and counting the salps, which were then discarded to be able to maintain the net contents in one bottle. A standard protocol really needs to be established regarding this issue for future legs.

EQUIPMENT & CONSUMABLES (e.g. filters, tubes, chemicals)

Biologist engineer (Celine Dimier). Inventory has been done with Steffi Kandels-Lewis at Abhu Dabi. Some filters and chemicals (47 mm 10um filter, aliquot frozen of glutaraldehyde) were not in sufficient amount because of protocols changes. I had to adapt by taking other size of filters (25 mm

diameter instead of 47mm) or doing aliquots by myself. Lugol was sufficient until Mumbai but will not until Malé.

SAMPLE STORAGE

Biologist engineer (Celine Dimier). The fridge could be too small or the sample storage until Male. One tank of LN2 is already full of samples. We also put samples in the second tank. To keep free space until Male in the LN2 tank, we were forced to store HPLC samples in the freezer, instead of keeping them in LN2.

METADATA & DATA

Oceanography engineer (Marc Picheral). Daily TSG files were sent to the datacenter during all leg. All CTD files were also daily processed, checked and transmitted to Coriolis. Backup performed on a daily base for all oceano instruments deployed on deck or managed in the DryLab.

OTHER

Oceanography engineer (Marc Picheral). A depth transducer is highly requested to monitor the depth of instruments in real time and adjust the length of cable to reach the intended depth. The last Multinet worked well but we passed the starting depth of only 43m on 1500m of deployed cable !