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Shipboard Microbiologist on IODP Drilling Vessel Chikyu, Expedition 370

From September 13th to November 11th 2016 I had the pleasure to contribute to IODP expedition 370 "T-Limit of the Deep Biosphere off Muroto" sailing aboard the Japanese drilling ship Chikyu. The main goal of the expedition was to determine until where the biosphere extends in the deep hot sediments off the Muroto Peninsula.

My main contribution to the achievement of the goal as shipboard microbiologist was to monitor contamination by drilling fluid in the cores, using the chemical tracer perfluoromethylcyclohexane (PFC). Secondly, I cut whole-round cores, a lot of them, and I was part of the shavers' team. I will explain in a moment all these processes.

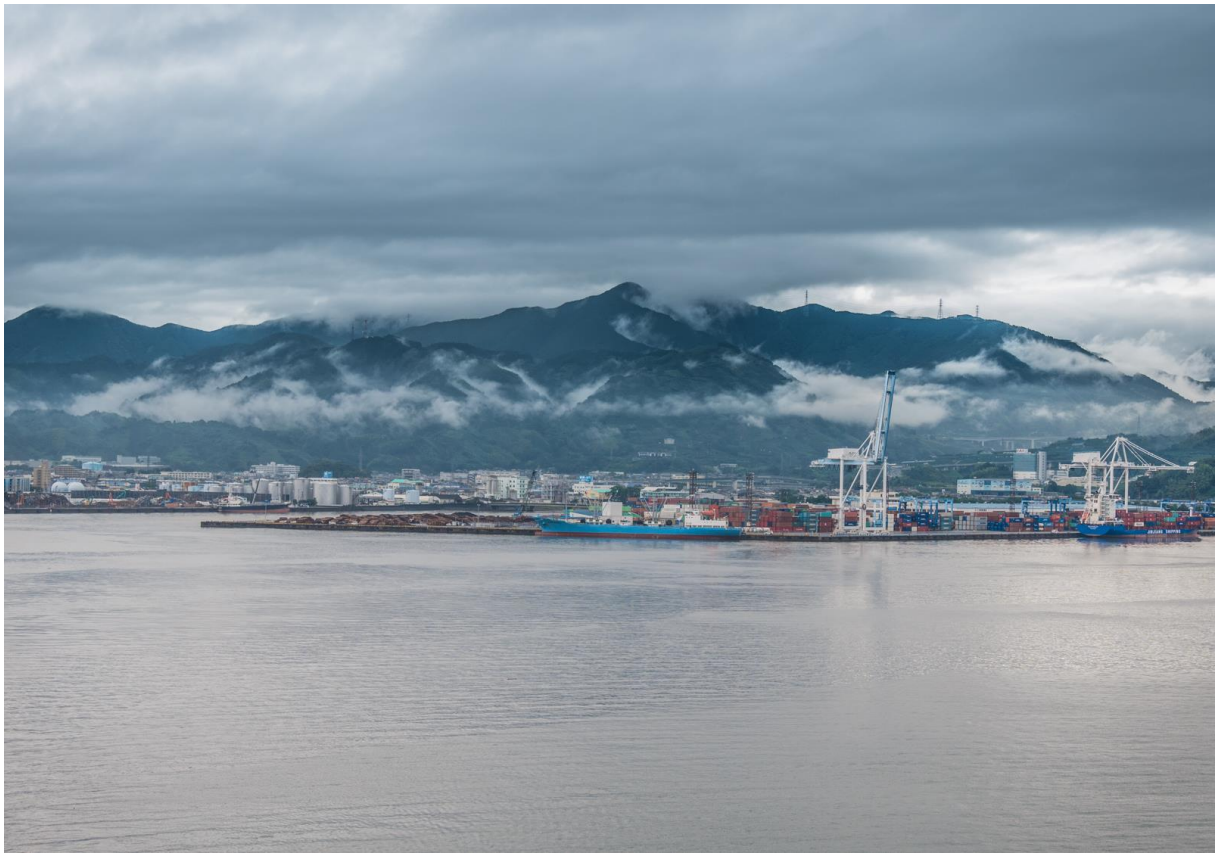

Figure 1. Clouds over Shizuoka port on the departure's day – September 13th 2016.

Why care about deep biosphere in first place?

Earth surface is covered by more than 70% of water, and the sediments at his bottom can be thick as much as 20 km! This makes it one of the biggest environment on Earth. In the last 20 years we realized that these sediments are not just barren as previously assumed but harbor a complex and fairly big microbial community - was estimated that subseafloors globally contains 2.9^29 cells, that corresponds to about 0.6% of Earth total living biomass. Despite tremendous advances in the study of microbial biogeography in subsurface marine sediments of the last two decades, still is unclear what is the extent of this biosphere, what are the environmental controls that are acting on these communities and what are the metabolic processes of the majority of subsurface microbes. As if this was not already exciting

enough to study microbes at the bottom of the oceans, it has to be considered that Ocean are the Earth's main Carbon sink, and that slowly this Carbon will eventually sink and be sequestered in sediments for millions of years. Microorganism inhabiting those sediments via organic matter remineralization and methanogenesis release CO_2 and CH_4, two important greenhouse molecules. In order to better understand Earth's Carbon budget and climate is thus crucial to characterize the processes happening at the bottom of the Oceans.

Why was selected this location?

The sediments off Shikoku Island have been previously investigated by several expeditions - in particular ODP expedition 190 drilled a site in close proximity of C0023, the site selected for expedition 370 – indicating C0023 as a promising site to study microbial communities in sediments characterized by high heat fluxes. The sediment column is about 1.2 km high, and present a thermal gradient penetrating into the sediments: at the surface, the temperature is only few °C but at the bottom sediment, where the Earth crust start, the temperature was estimated to be around 130°C.

Figure 2. Location of site C0023.

Contamination

Since the main goal of the expedition was to determine until where are microbes thriving in the deep hot sediments, crucial was to be able to estimate contamination, in order to not get excited for some false positive results! An intrusion of just few drop of seawater would completely alter the results, nullifying all our efforts. Taking pristine samples from 1 km below the surface of the sediments, with a water depth of almost 5 km using a big drill is something extremely challenging, but turned out to be possible. A series of geochemical, microbiological and chemical analysis were devoted just to guarantee that only pristine samples were used for contamination sensitive analysis.

As soon as the core was retrieved on board was cut into 1.5 meters sections, and those were scanned using 3D X-ray CT scan to look for internal micro-fractures (Fig 2). This is the same technology that is used to scan your knees if you injury yourself! After that pieces of core that were determined to be intact by X-ray scan were cut by the microbiology team into personal and community samples.

Figure 3. Technician Soichi Moriya preparing a fresh core for the 3D X-ray CT scan.

To guarantee that the most contamination sensitive samples were not contaminated by drilling fluids (a mixture of seawater and viscosifiers that help the drilling process and that are pumped down into the borehole) were subsampled and checked for presence of PFC, a chemical tracer that was added to the drilling fluid. Other geochemical and molecular analysis (sulphate profiles and DNA sequencing respectively) were also used to estimate contamination.

The last task of microbiologist was to shave cores designated to the most contamination sensitive procedure (cultivation, enzymatic assays). This was done in an anaerobic glove box to remove any mud on the surface of the core that was introduced by drilling. The shaving was done with Japanese ceramic knives.

Samples were then carefully packed: either vacuumed and deep freezed or stored at 4°C and shipped by helicopter to Kochi Core Center, where the shore-based team was ready to process them. This was done in order to be able to start to perform time sensitive analysis with no delays, increasing potential success of cultivation and isolation of new microbial species.

More information on the analysis that were done onboard and onshore, and that will be performed in the coming months after the expedition are available on the preliminary report of the expedition:

http://publications.iodp.org/preliminary_report/370/

Life on Chikyu

Expeditions are extremely intense, and can be quite stressful, working more than 12 hours every day for weeks in an unusually limited environment. Probably also for this reason it was one of the most rewarding experiences of my life. Working together towards a shared goal make everyone striving to give their best and help each other. The feeling of cooperation that I felt onboard - and probably other freshman sailors experienced – was something I never experienced so strong on land.

Aside from work we had several ways in which we could relax and distract from work: gym and sauna were often used by the scientific party (and crew as well). The most waited moment of the day was the sunset (at least for the day shift) were everybody, if not processing a core, would have gathered on the helideck could to enjoy the breathtaking view of the sun sinking in the ocean.

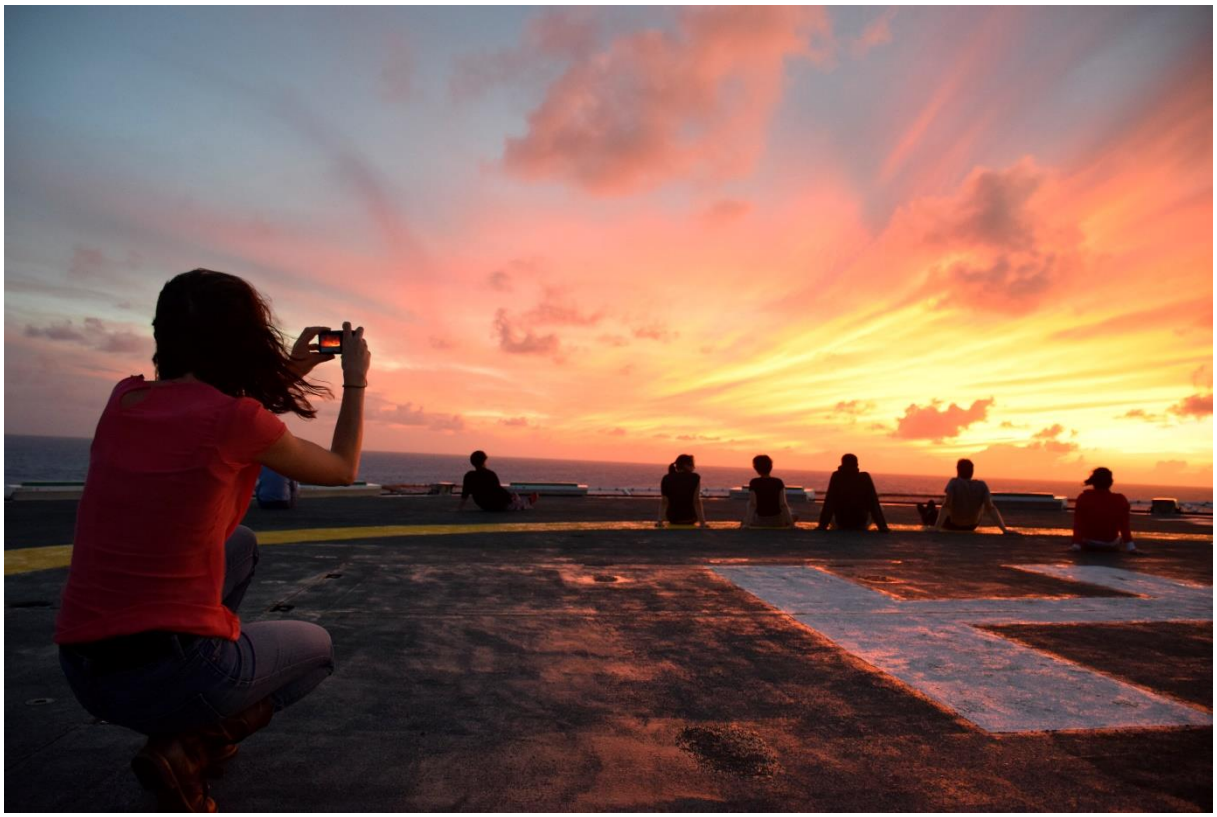

Figure 4. Stunning sunset from the helideck.

In the second part of the cruise, apart from cores and science in our heads there was room for only one thing: the ping-pong tournament. The tournament was organized by Masanori Kaneko, the ping-pong coach, and was held using official international ping-pong rules. Even participants who claimed to not care about the tournament were found training in secret, developing new shots and effects serves, and even planning their diet to perform better during the trainings. Just to mention, the final was won by the Japanese scientist Takehiro Hirose, one of the favorites from the beginning, now all-time best Chikyu player having won two tournaments on board.

Figure 5. Ping-Pong tournament's initial plan.

Figure 6. The apprentice (Florence Schubotz) VS the master (Arthur Spivack) competing for one spot in the semifinals.

Figure 7. Getting ready for the Sunday morning drill. Securing lifejackets in the proper way required much more dexterity than what the safety video wanted make to us believe.

Science goes on

The expedition ended in November 2016, but due to the challenging nature of the samples, and the huge amount of samples took (it was the IODP expedition that collected the highest number of samples for microbiology) sediments collected in IODP expedition 370 will be analyzed for years, by master students, PhDs, postdoc etc. This will hopefully leave a deep impact in our knowledge of subseafloor biosphere!

Figure 8. The ship base team is full of energy. Find Donald.