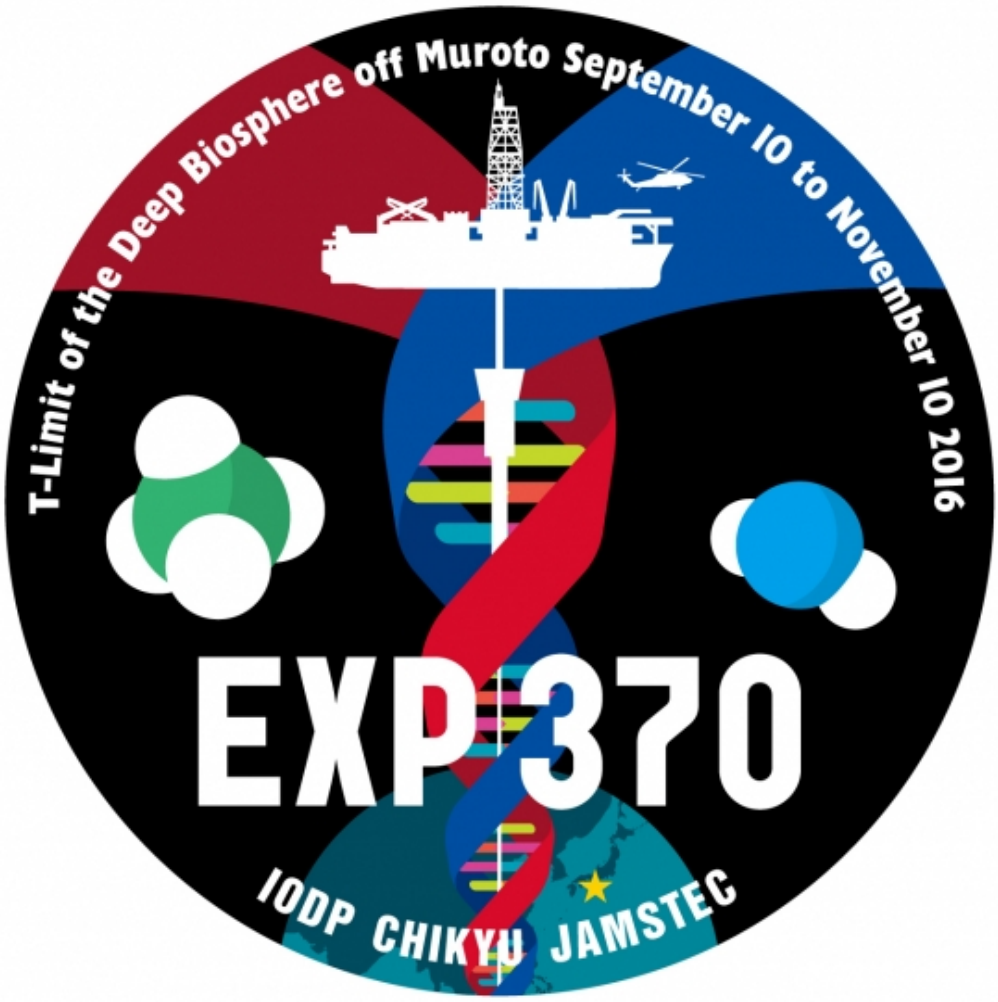

Abstracts and Session Plans for IODP 370 Post-Cruise Meeting

31st May to 3rd June 2018

Meeting supported by:

UK IODP INTERNATIONAL OCEAN DISCOVERY PROGRAM scientific ocean research drilling

Friday 1st June
0900-1700 Scientific Sessions

Part I: Impact of temperature on the abundance and activity of microbial life at Site C0023

0900 – 1030: Session I:

1. Final temperature model for Site C0023 (15 minutes)
   Takehiro Hirose & Masa Kinoshita: The shipboard temperature model and preliminary results of the temperature observatory

2. Abundance of Microorganisms at Site C0023 (5 x 15 minutes)

- Yuki Morono: Cell counts & high-pressure-high-temperature incubations, QA/QC
- Bernhard Viehweger: Endospore distribution inferred from the biomarker dipicolinic acid (DPA)
- Casey Hubert: Update on attempts to detect thermophilic spores
- Donald Pan: Virus enumeration
- Tatsuhiko Hoshino: On the endeavor to extract in situ microbial community structure

1030 – 1100: Break

1100 – 1200: Session II

3. Microbial Activity at Site C0023 (45 + 15 minutes)

- Radio Tracer Team (45 minutes, one combined talk)
  Tina Treude, Jens Kallmeyer, Felix Beulig, Rishi Adhikari, Clemens Glombitza, Florian Schubert: Radiotracer incubations to determine microbial methanogenesis, methane oxidation, sulfate reduction, and hydrogenase activity
- Rachel Harris (15 minutes) Stable isotopic evidence of high-pressure, high-temperature anaerobic methane oxidation

4. Concluding discussion of session I and II (15 minutes)

1200 – 1300: Lunch

1300 – 1730: Session III, including Video Chat with David and 30 minutes coffee break

5. Biogeochemistry at Site C0023 (7 x 15 minutes)

- Masanori Kaneko: Distribution of methanogenic coenzyme F430 at Site C0023
- Man-Yin Tsang: Porewater and mineral sulfur species
- Male Köster & Susann Henkel: Biotic iron reduction & trace elements in interstitial waters and bulk sediments
- Kira Homola: Subsurface nitrogen cycling controls and implications
- Art Spivack: Coupled transport and thermodynamic controls on deep biosphere activity
- Akira Ijiri: Stable isotope geochemistry of pore waters
- Shuhei Ono & Video Chat with David: Clumped isotopes and gas extractor

15:15 – 16:00 Coffee Break (15:15 – 16:00; including 15 minutes buffer for video chat)

6. Microbe-mineral interactions at Site C0023 (2 x 15 minutes)
- Kyle Metcalf: Mineral-associated microbial ecology of marine sediments
- Jinwook Kim: Microbe-clay mineral interaction at various temperatures

7. Culturing of microbes from Site C0023 (2 x 15 minutes)
- Hiro Imachi: Cultivation of decollment life with continuous-flow bioreactors
- Maija Raudsepp: Adventures in culturing microbes from the deep biosphere

8. Summary and concluding discussion of Part I (Session I-III) (30 minutes)

Venue: MacRobert Building

Free Evening

Saturday 2nd June
0900-1200 Scientific Sessions

Part II: Investigate other factors that potentially influence microbial life (or vice versa) at Site C0023

0900 – 1015: Session IV

9. Lithology, tectonics and fluid flow at Site C0023 (5 x 15 minutes)
- Yuzuru Yamamoto:
  Structural characteristics of the décollement zone and underthrust sediments in the Nankai accretionary prism: Geologic architectures in the Site C0023, IODP Expedition 370
- Yasuhiro Yamada: Initiation process of frontal thrust at accretionary prism
- Takehiro Hirose: Excess fluid pressure development beneath the décollement
- Nana Kamia: Paleo-thermal anomaly along with the decollement off the Cape of Muroto, Japan: controlled by subduction
- Stephen Bowden: Getting Into Hot-Water in the Underthrust Sediments of Nankai Accretionary Complex
- Satoshi Tonai: A new method for measurement of geological core sample quality using X-ray Computed Tomography data and its application of IODP Expedition 370

1015 – 1045: Coffee Break

1045 – 1215: Session V

10. Impact of temperature and life on mineralogy (4 x 15 minutes)
- Myriam Kars: Effects of high temperature and a deep SMT on rock magnetic properties
- Natsumi Okutsu: Progress toward understanding the characteristics and sedimentary system of silt-turbidites of IODP Site C0023
- Kiho Yang: Application of EELS on the microbial mineral alteration
- Oliver Helten & Florence Schubotz: Abiotic substrate generation from recalcitrant organic matter at elevated temperatures

11. Concluding discussion of Part I and II (30 minutes)

Venue: MacRobert Building

1215 - 1400 Tours of Old Aberdeen (incl. group photo)

Part III: Publication strategies

1400 – 1530 Group Discussion #1
1530 - 1700 Group Discussion #2

Venue: MacRobert Building

Sunday 3rd June
0900-1100 Wrap up Sessions
10 mins summary from groups
Wrap up by co-chiefs and lead proponent
Venue: MacRobert Building

The end
1300 Space arranged for afternoon meetings if needed

Part I: Impact of temperature on the abundance and activity of microbial life at Site C0023
0900 – 1030: Session I:

12. Final temperature model for Site C0023 (15 minutes)
    Takehiro Hirose & Masa Kinoshita: The shipboard temperature model and preliminary results of the temperature observatory

13. Abundance of Microorganisms at Site C0023 (5 x 15 minutes)

- Yuki Morono: Cell counts & high-pressure-high-temperature incubations, QA/QC
- Bernhard Viehweger: Endospore distribution inferred from the biomarker dipicolinic acid (DPA)
- Casey Hubert: Update on attempts to detect thermophilic spores
- Donald Pan: Virus enumeration
- Tatsuhiko Hoshino: On the endeavor to extract in situ microbial community structure

Endospore distribution in the Nankai Trough deep biosphere - Insights from IODP Expedition 370

B. Viehweger*, N. Gajendra, L. Warmer, V.B. Heuer and K.-U. Hinrichs

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Expedition 370 of the International Ocean Discovery Program (IODP) established Site C0023 in the Nankai Trough off Cape Muroto, Japan, to investigate the upper temperature limit of microbial life in deep subseafloor sediments. Temperatures ranged from ~30 °C at 200 mbsf to ~120 °C at 1200 mbsf covering mesophilic to hyperthermophilic growth conditions of microbes known from cultivation based studies. With increasing burial depth, sediments heat up progressively and nutrients become increasingly depleted. The formation of highly resistant endspores by bacteria of the phylum Firmicutes enables them to survive under these otherwise lethal conditions. Thus, the investigation of endospores can provide important insights into the limits of life in the deep biosphere. To quantify endospores we used the biomarker dipicolinic acid (DPA) specific for intact endospores. In total, we extracted 1 g of each sediment sample with a newly developed protocol to achieve a higher extraction efficiency. To maximize data quality, each extraction set consisted of five randomly chosen samples in duplicates and two extraction blanks. We analyzed DPA extracts by high performance liquid chromatography coupled to a fluorescence detector following a dedicated method [1] and converted DPA into endospore concentrations. Here we will present a high-resolution record of endospore abundance in the Nankai Trough deep biosphere setting covering temperatures from 30 to ~120 °C. We will discuss our findings in the context of lithology, sediment age, and the preservation potential of the biomarker DPA in the up to 16 Ma old sediments.
[1] Fichtel (2007), J. Microbiol. Meth. 70, 319-327."

Thermophilic Spores

Margaret Cramm, Carmen Li & Casey Hubert

Email: chubert@ucalgary.ca
Address: University of Calgary

An update on attempts to detect thermophilic spores in the IODP 370 samples.

Development of an improved virus enumeration method for TLIMIT (and other) sediments

Donald Pan, Yuki Morono, Fumio Inagaki, Ken Takai

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The extent to which life inhabits the deep subsurface is still a major question of subsurface microbiology, especially in low energy environments where low concentrations of cells challenge current limits of detection. Recent technical advances have improved the recovery of cells from deep subseafloor sediments making it possible to enumerate cells at extremely low concentration. Viruses have also been observed in subseafloor sediments, however the limits of the subsurface virosphere are not well constrained. Viruses are the most abundant biological entities on Earth's surface and are major components of biogeochemical cycles, however current limits of detection are insufficient for investigating the limits of the virosphere. Improvements in virus recovery from subseafloor sediment are still necessary to extend the limit of detection of viruses in subseafloor sediment. Here, we developed an improved method for the extraction of viruses from subseafloor sediment using low-speed Nycodenz density gradient centrifugation to remove interference from non-biological particles. The new method resulted in an increase in extraction efficiency from deep subseafloor sediments compared to conventional methods. Applying this method, we find that previously published virus abundances from subseafloor sediments dramatically underestimate subsurface viral abundances by as much as 2-3 orders of magnitude. This method enables us to detect viruses in sediments that would have previously been below limits of detection, thus potentially expanding the known boundaries of the deep virosphere.

An endeavour to extract *in-situ* microbial community structure

Tatsuhiko Hoshino and 370 Expedition Scientists

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During the expedition, we extracted DNA from every core and obtained a sequence library containing 16S rRNA gene sequences from more than 100 sediment samples. Unexpectedly low biomass even in the shallow sediment hamper to determine in situ microbial community because the real signal was probably hidden behind contaminated DNA. In silico analysis of the sequence library obtained during the expedition indicated that a substantial number of sequences could from contaminated DNA. Because no archaeal sequence was found in negative controls, one solution is targeting archaeal 16S rRNA gene by using domain specific primers. However, archaea-specific PCR resulted in no amplification product from deeper sediments, indicating need to re-extract DNA. To determine in situ microbial community structure, we are now trying to extract high-quality DNA by using large volume ~100 g of sediment containing more than 100 cells/cc for the extraction. In the presentation, I will show our procedure to obtain high quality of DNA from very low biomass samples and hopefully discuss relation between microbial community structure and geochemical data of sediment.

1100 – 1200: Session II

14. Microbial Activity at Site C0023 (45 + 15 minutes)

- Radio Tracer Team (45 minutes, one combined talk)
  Tina Treude,
  Jens Kallmeyer,
  Felix Beulig,
  Rishi Adhikari,
  Clemens Glombitza,
  Florian Schubert:
  Radiotracer incubations to determine microbial methanogenesis, methane oxidation, sulfate reduction, and hydrogenase activity

- Rachel Harris (15 minutes) Stable isotopic evidence of high-pressure, high-temperature anaerobic methane oxidation

Exploring microbial sulphate reduction under high temperature and pressure; results of a pilot study on samples from IODP Exp. 370

Florian Schubert, Jens Kallmeyer, Tina Treude

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Helmholtz-Zentrum Potsdam - Deutsches GeoForschungsZentrum GFZ, Telegrafenberg, 14473 Potsdam, Germany

Sulphate reduction is the quantitatively most important process in the anaerobic degradation of organic matter in marine sediments. Data from hydrocarbon reservoirs suggested that microbial activity in non-hydrothermal sedimentary systems ceases above 80 °C. Due to the high heat flow in the Nankai Trough area the geothermal gradient is high enough (ca. 100 °C km-1) to allow exploration of the putative biotic fringe at a relatively shallow depth, but with high resolution of the temperature gradient. Each of the most important microbial turnover processes (methanogenesis, anaerobic oxidation of methane, hydrogenase enzyme activity, sulphate reduction) will be measured by radiotracer incubations by a different group that has specialized in this kind of analysis. Incubations for sulphate reduction rates were carried out close to in-situ temperature but at atmospheric pressure. Like at ODP Leg 190, Site 1174, which was drilled in close proximity to Exp. 370 site C0023, sulphate reduction could be detected down to over 1000 mbsf. Results from incubations without substrate addition show a steep decline at the transition from a mesophilic to a thermophilic temperature regime (40-60 ° C), but activity remains low but detectable throughout the core. Additions of volatile fatty acids and methane increased activity in the upper ca. 400 mbsf by several orders of magnitude but very little below. However, methane addition appeared to cause an increase in activity in the deepest sample. Our results show that sulfate reduction proceeds at temperatures up to at least 95 ° C, expanding the range of the habitable subsurface realm.

Hydrogenase enzyme activity

Rishi Ram Adhikari, Verena Heuer, Kai-Uwe Hinrichs and IODP Exp. 370 Scientists

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The subsurface harbours complex microbial ecosystems, capable of many metabolic processes that are closely coupled to their biogeochemical environment. Given a number of simultaneously occurring processes, it is always a major challenge to quantitatively study such biosphere. Here we present hydrogenase enzyme activity assay to investigate potential microbial activity based on hydrogen turnover mechanism. Hydrogenases are a large and very diverse family of enzymes expressed by all microorganisms that utilize elemental hydrogen in their metabolic pathway, irrespective of whether they produce or consume hydrogen or use it as a metabolic intermediate. Because hydrogenases only occur intracellularly, any detected activity can be related to intact cells that are most probably metabolically active. Potential hydrogenase enzyme activity was measured in sediment samples from IODP Expedition 370. Hydrogenase enzyme activity was above detection limit at most of the samples. Volumetric hydrogenase activity at Unit II remained <5 µmol H_2/g/d, whereas the activity at Unit III is mostly >10 µmol H_2/g/d. Highest activity of ca. 50 µmol H_2/g/d were observed at the uppermost part of the Unit IV which continuously decreased over depth with some scatter. Cell specific activity could provide more insights into the potential hydrogen turnover rate activity.

Stable isotopic evidence of high-pressure, high-temperature anaerobic methane oxidation in sub-seafloor sediments from IODP 370 site C0023A: insights and investigations from a one-year incubation experiment

Rachel L. Harris, Douglas Bartlett, Tatsuhiko Hoshino, Alexander W. Byrnes, Keeley M. Walsh, Chui Y. M. Lau, and Tullis C. Onstott

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The anaerobic oxidation of methane (AOM) is a microbially-mediated process that plays a significant role in controlling the net atmospheric flux of methane (CH4) from the oceans. Anaerobic methanotrophic archaea (ANMEs) can mediate AOM by oxidizing a variety of electron acceptors either independently or in syntrophy with bacteria. ANMEs heretofore have only been documented to survive up to 50 °C by coupling CH_4 oxidation to sulfate reduction. We report evidence of the biological AOM in high-pressure, high-temperature enrichments whose inoculum does not appear to contain canonical ANMEs. Sediments from 9 whole core rounds from 257 - 865 m below seafloor (mbsf) were collected during IODP 370 in the Nankai Trough and incubated anaerobically for 350 days at 40 MPa and 40 – 80 °C to simulate in situ conditions. Incubations contained sulfate-free artificial seawater, a 2:98 $^{13}CH_4$:N_2 headspace, and one of the following electron acceptors: $SO4^{2-}$, NO^{3-}, NO^{2-}, Fe^{3+}, Mn^{4+}, plus autoclaved kill, no added oxidant, and background heterotrophy controls. Stable isotope analysis of dissolved inorganic carbon (DIC) suggests that AOM is occurring in enrichments for all investigated depths up to 80°C, with the highest rate measured at 14 days in Fe-dependent AOM enrichments from 257 mbsf (1.25 pmol ± 0.04 $^{13}CO_2$ cm^{-3} d^{-1} relative to kill). This talk will describe trends observed in rates and modes of AOM recorded over the past year, as well as current efforts underway in elucidating the organisms implicated in these pathways.

1300 – 1730: Session III, including Video Chat with David and 30 minutes coffee break

15. Biogeochemistry at Site C0023 (7 x 15 minutes)

- Masanori Kaneko: Distribution of methanogenic coenzyme F430 at Site C0023
- Man-Yin Tsang: Porewater and mineral sulfur species
- Male Köster & Susann Henkel: Biotic iron reduction & trace elements in interstitial waters and bulk sediments
- Kira Homola: Subsurface nitrogen cycling controls and implications
- Art Spivack: Coupled transport and thermodynamic controls on deep biosphere activity
- Akira Ijiri: Stable isotope geochemistry of pore waters
- Shuhei Ono & Video Chat with David: Clumped isotopes and gas extractor

Distribution of methanogenic coenzyme F430 at Site C0023 off Muroto

Masanori Kaneko and Expedition 370 Scientists

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Coenzyme F430 is a hydrocorphinoid Ni complex which acts an active site on methyl coenzyme M reductase and catalyzes the last step of methanogenesis. Since this last step is common in all methanogenic pathways, coenzyme F430 can be a good biomarker for methanogenesis. In this study, coenzyme F430 analysis was applied into sediment core collected during IODP Expedition 370 to investigate distribution and activities of methanogens and limits of methanogenesis in a high heat flow region of Nankai Trough. So far, coenzyme F430 was analyzed from about 30 sediment depths: down to 600 mbsf with low resolution, below 600 mbsf with relatively high resolution. Concentrations of coenzyme F430 were between 0.9 and 3.6 fmol above 600 mbsf. Coenzyme F430 was detected in all sediment samples below 600 mbsf in very low concentrations but it was hard to determine the precise concentration due to similar levels of contamination. In this talk, I will show the results of coenzyme F430 analysis and assessment of contamination.

Porewater and mineral sulfate at the Nankai Accretionary Complex (IODP Exp. 370 Site C0023)

Man-Yin Tsang, Stephen Bowden, Kiho Yang, Ulrich Wortmann, Verena B. Heuer, Fumio Inagaki, , Yuki Morono, and Expedition 370 Scientists

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Here we report the $\delta^{34}S$ of dissolved sulfate in porewater at Site C0023 for quality assurance and recognising microbial sulfate reduction. From 190 to 450 metres below seafloor (mbsf) where porewater water sulfate is of low concentrations (<1 mM), measured sulfate $\delta^{34}S$ are at seawater values, suggesting seawater as the major contributor to the sulfate measured. From 680 to 920 mbsf, δ34S of porewater sulfate significantly increases. In conjunction with the expense of methane, the increase in δ34S suggests microbial sulfate reduction / anaerobic oxidation of methane in these sediments across the décollement zone. Further down in the sediment column, paleo-sulfate and depleted anhydrite could both contribute to the measured $\delta^{34}S$. During Expedition 370, barite and anhydrite were described during visual core description as veins or euhedral minerals below the décollement zone. Their $\delta^{34}S$ are also measured in order to understand their origins and their potential effects to the profile of porewater sulfate. Such vein and euhedral barite has $\delta^{34}S$ significantly more enriched than porewater sulfate as well as the fine barite in background mudrock. This suggests that either the veins were formed with external, enriched fluid, or faults and joints in sediments create refuges in which sulfate reduction might continue to occur.

Examining the role of biotic iron reduction as a life-sustaining process at the potential temperature limit of the deep subseafloor biosphere (RESPIRE), IODP Expedition 370: Trace elements in interstitial water and bulk sediment composition at site C0023A

M. Köster, S. Henkel, A. Spivack, K. Homola, J. Sauvage, V.B. Heuer, F. Inagaki, Y. Morono and Exp. 370 scientists

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Iron reduction is considered one of the most ancient forms of microbial respiration. This and the observation that iron reducers can grow under high temperature and pressure conditions suggest that Fe(III)-reducing microbes may represent a significant part of the deep biosphere. We give an overview of the RESPIRE project, in which we aim at using stable Fe isotopes of dissolved and reactive solid Fe to discriminate microbial and abiotic drivers of Fe(III) reduction in deep subseafloor sediments of site C0023A. As a first step of the project, the total element composition (e.g., Al, Ba, Ca, Fe, Mn, S, Ti) of solid phase samples was determined via acid digestion. The Fe content fluctuates between 3.9 and 4.6 wt% from 400 down to 670 mbsf, coinciding with the occurrence of volcanic ash layers in Subunit IIC and the Upper Shikoku Basin facies. Further below, total Fe is relatively constant at around 4.4 wt%. An increase in total Fe up to 5.1 wt% occurs between 1033 and 1090 mbsf in the Lower Shikoku Basin facies. The Mn content generally increases downcore, which is in agreement with the progressive occurrence of Mn carbonates at depth. The Ca content strongly varies between 0.7 and 7 wt% with highest contents in the Upper Shikoku Basin and the top of the Lower Shikoku Basin facies. Trace element data were gained for interstitial water by mass spectrometry and will be presented.

Subsurface Nitrogen Cycling Controls and Implications

Kira Homola, A. J. Spivack, C. Buchwald, S. D. Wankel, R. S. Robinson, C. N. Charoenpong, E. R. Estes, R. W. Murray

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The subseafloor microbial nitrogen (N) cycle influences the availability and speciation of dissolved N through a variety of redox processes. Isotopic measurements of porewater ammonium (NH_4^+), nitrite (NO_2^-), and nitrate (NO_3^-) can be combined with concentration and solid phase geochemical data to trace the cycling of these compounds through the subsurface. Constraining the subseafloor isotopic nitrogen cycle could provide insights ranging from microbial energy limitation to the evolution of nitrogen across Earth's reservoirs and geologic history. We present results from two recent studies of porewater nitrogen isotope profiles; KN223 Site 3 (Researcher Ridge, North Atlantic) and IODP Site C0223 (Nankai Trough, Japan).We use porewater concentrations and stable N and O isotope measurements of nitrate and nitrite to constrain rates of nitrogen cycling processes over a 34 m profile from the deep North Atlantic spanning fully oxic to anoxic conditions. Using a 1D reaction-diffusion model to predict the distribution of nitrogen cycling rates, results converge on two distinct scenarios: 1) an exceptionally high degree of coupling between nitrite oxidation and nitrate reduction near the top of the anoxic zone or 2) an unusually large N isotope effect (~60‰) for nitrate reduction that is decoupled from the corresponding O isotope effect, which could be explained by enzyme-level interconversion of nitrite and nitrate.
IDOP 370 porewater was measured for ammonium concentration shipboard and aliquots were frozen at -80C for postcruise nitrogen isotope analysis. Preliminary total dissolved nitrogen (TDN) isotopes span a range from 2.5 to 6.2 ‰ from 250 to 1075 mbsf, with features of interest corresponding to the sulfate-methane transition zone (SMTZ), decollement, and deep sulfate minimum.

Stable isotope geochemistry of pore waters in the Nankai accretionary prism off Muroto

Akira Ijiri, Fumio Inagaki

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I analyzed the stable oxygen and hydrogen isotopic compositions of pore water. Between 600 mbsf and 1000 mbsf, $d^{18}O$ and dD values and Cl showed ^{18}O enrichment and D-depletion in proportion to Cl^- depletion, suggesting the mixing of freshwater derived by clay mineral dehydration.

15:15 – 16:00 Coffee Break (15:15 – 16:00; including 15 minutes buffer for video chat)

16. Microbe-mineral interactions at Site C0023 (2 x 15 minutes)

- Kyle Metcalf: Mineral-associated microbial ecology of marine sediments
- Jinwook Kim: Microbe-clay mineral interaction at various temperatures

17. Culturing of microbes from Site C0023 (2 x 15 minutes)

- Hiro Imachi: Cultivation of decollment life with continuous-flow bioreactors
- Maija Raudsepp: Adventures in culturing microbes from the deep biosphere

Cultivation of décollement life with continuous-flow bioreactors.

Hiroyuki Imachi

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In microbiology and its related fields, cultivation and isolation of microorganisms is one of the important challenges because cultivation/isolation of microbes is the most convicting and direct way to know exactly how they make a living. However, many microorganisms have been highly resistant to in vitro cultivation such as conventional batch-type cultivation technique. To overcome this situation, I use a continuous-flow bioreactor cultivation technique called down-flow hanging sponge (DHS) reactor for cultivation of subseafloor microorganisms. In this study, I applied DHS bioreactor technique to cultivate hyperthermophilic, anaerobic microorganisms associated with methane-production and methane-oxidation from the Nankai Trough subduction zone. For cultivation of methanogenic microbial community, I anaerobically incubated samples collected from the décollement zone in a DHS reactor and provided anaerobic medium supplemented with acetate, glucose, yeast extract and amino acids. To cultivate anaerobic oxidation of methane (AOM) microbial community, I used 48R-1 core sample obtained from a sulfate-methane transition zone and incubated the samples in a DHS reactor with methane and anaerobic synthetic seawater based medium congaing sulfate and thiosulfate as electron acceptors. For the AOM bioreactor, I also supplied a trace amount of ethane, propane and butane in addition to methane. Both of the DHS bioreactors were operated at 85 °C for about 9 months. During the reactor operation, I monitored methane concentrations in the head space of the reactors and observed microbial cells in effluent samples from the bioreactors. However, unfortunately, no methane production and microbial cells were observed. 16S rRNA gene-tag sequencing data for the incubated samples also indicated that no hyperthermophiles grew in the bioreactors. These results suggest the difficulties in cultivation of microorganisms that live at the edge of habitable zone.

Adventures in Culturing Microbes from the Deep Biosphere

Maija Raudsepp and Gordon Southam

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During IODP Exp. 370 detailed organic and inorganic geochemical profiles were produced. Changes in chemical constituents with depth suggest possible changes in microbial metabolic processes. The purpose of this study was to enrich mesophilic and thermophilic methanogens, iron reducing microorganisms and other novel microbial groups from deep subseafloor sediment and to study the interaction between cells and the sedimentary minerals. There are currently 62 on-going incubations of sediment under 13 different medium conditions and three different temperatures (30 °C, 50 °C and 70 °C). On-going monitoring with fluorescence microscopy has been challenging but several, putative bacteria have been observed in some of the sediment incubations. In some low biomass environments, a higher rate of cellular growth occurs only after reaching a critical cell number, i.e., there is a long delay before growth starts. Currently, only one high biomass enrichment culture has grown from collected the deep subseafloor sediment. This culture is sourced from 470 mbsf and grows on a mixture of lactate and acetate at 50 °C and DNA from this culture has been submitted for 16S rRNA gene sequencing. With continued incubations, further enrichments are expected. As most thermophilic microbial isolates have been obtained from hydrothermal vent environments, the enrichment of hot and deep subseafloor microorganisms may result in novel microbial species.

Part II: Investigate other factors that potentially influence microbial life (or vice versa) at Site C0023

0900 – 1015: Session IV

18. Lithology, tectonics and fluid flow at Site C0023 (5 x 15 minutes)

- Yuzuru Yamamoto:
  Structural characteristics of the décollement zone and underthrust sediments in the Nankai accretionary prism: Geologic architectures in the Site C0023, IODP Expedition 370
- Takehiro Hirose: Excess fluid pressure development beneath the décollement
- Yasuhiro Yamada
- Nana Kamia
- Stephen Bowden
- Satoshi Tonai

Structural characteristics of the décollement zone and underthrust sediments in the Nankai accretionary prism: Geologic architectures in the Site C0023, IODP Expedition 370

Yuzuru Yamamoto*, Yuhji Yamamoto, Natsumi Okutsu, Yasuhiro Yamada, Stephen Bowden, Satoshi Tonai, Kiho Yang, Man-Yin Tsang, Takehiro Hirose, Nana Kamiya, and Expedition 370 Scientists

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Expedition 370 penetrated the accretionary prism, plate boundary décollement zone, and underthrust sediment and touched the basement basalt on the Philippine Sea Plate. Compared to the legacy sites (Sites 808 and 1174), the décollement zone is characterized by weak and intermittent negative reflectors in the seismic profile. Onboard physical properties, e.g. porosity and P-wave velocity data, indeed show the smaller gaps at the top of the décollement zone. The Décollement zone in Site C0023 represented the weaker and non-localized deformation zone comprised of alternating zone of ~1 m thick phacoidal deformation zones and ~a few 10 m of intact intervals in the Site C0023. Many normal faults striking parallel to the trench were identified just below the décollement zone, which is indicative of non-localized deformations along the décollement zone. Many of these faults were accompanied with calcite and sulphate mineral veins (anhydrite and barite), indicative of high-temperature fluid migration just above the ridge-spreading center. Based on the paleomagnetic restoration of structure to the geologic coordinate, attitudes of the bedding and fault planes in the Site C0023 are controlled by two factors: 1) subduction/accretion producing the trench-parallel bedding strikes and trench-perpendicular principal stress and 2) ridge spreading that produces ridge-parallel bedding and vein strikes. The former developed in the accretionary prism and the upper part of the underthrust sediment (<900 mbsf), whereas the latter occurs in the lower part (>900 mbsf). These tectonic variations might affect fluid migration pathways.

Excess fluid pressure development beneath the décollement at IODP Site C0023

T. Hirose, Y. Hamada, N. Kamiya, Y. Yamamoto, V. Heuer, F. Inagaki, Y. Morono, Y. Kubo, and Expedition 370 Scientists

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We report (1) estimate of the pore pressure developed within the underthrust sediments in the Nankai Trough off Cape Muroto, and (2) equivalent strength along the drilling hole, using the shipboard data obtained at Site C0023 during IODP Expedition 370.

Initiation process of frontal thrust at accretionary prism

Yasuhiro Yamada, Yuzuru Yamamoto, IODP 370 Scientists

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Deformation front of thrust belts conserves the features related to the initial process of shortening deformation. According to a series of 2D (cross-sectional) analog experiments by Dotare et al (2016), a number of ""weak shear bands"" with spatial and temporal variations may be produced prior to a thrust initiation, and these shear bands may show a nonuniform distribution around the new thrust fault above the decollement surface. IODP 370 was drilled at a deformation front of the Nankai Trough accretional wedge, a well-known typical thrust belt generated by a plate subduction. After the expedition was conducted, all of the core -CT scan data was examined to identify fracture distributions. The result shows a pattern similar to the prediction based on the analogue model suggesting that the fracture systems in the cores may be consisting shear bands in the prism and formed in a similar manner with the model. This strongly suggests that possible fluid flow coming from the deep may not be continuous but rather intermittent through the plate boundary fault then these fracture surfaces.

Paleo-thermal anomaly along with the decollement off the Cape of Muroto, Japan: controlled by subduction

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Vitrinite reflectance analysis has been used to estimate the paleo-maximum temperature of IODP 370 site C0023 located off Cape of Muroto. Vitrinite reflectance increases from 0.2 to 0.6 % from 200 to 700 mbsf. At locations deeper than 700 mbsf, the reflectance is almost constantly around 0.6 %. For durations of heating ranging of 400, 4000, 40000 and 400000 years the paleo-maximum temperature was estimated using the Sweeney and Burnham (1990) model of vitrinite reflectance. Within the depth range 200 to 750 mbsf temperatures are within 40~140 ˚C but could be greater than 140 ˚C at a depth of 750 mbsf or deeper. The paleotemperatures estimated from vitrinite reflectance are higher than the present temperature calculated from APCT3 data and thermal conductivity modelling, which suggests that thermal regimes were hotter in the past. Paleaotemperature data also indicate a high temperature zone within the vicinity of the décollement zone, indicating the possibility that the décollement zone has served as a loci of heating in addition to heat flow from the basement. This observation aggress with other studies that suggested advection of high temperature fluids along the décollement, and observations that of barite deposited from high temperature fluids.

Getting Into Hot-Water in the Underthrust Sediments of Nankai Accretionary Complex

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Using hydrothermal mineral assemblages, fluid inclusion homogenisation temperatures and hopane biomarkers it is possible to investigate thermal regimes within the Underthrust sediments in the Nankai Accretionary Complex. These data suggest two things; foremost that the temperature within the underthrust sediments at the present day may not reflect temperatures during the past, and secondly that thermal regimes within the underthrust sediments may not simply increase linearly with depth; that is assumptions about a given thermal regime at one depth may not be applied at another even if it is only a short distance away. This can be seen in models of the mineralisation halos found within sediments. These show that the heating required to precipitate minerals would create an aureole that extended only 10's of cm from a point, and at temperatures of 180 degrees centigrade this heating would be too short lived to generate methane from kerogen by catagenesis. Based on industrially applied sterilisation curves for thermopiles, used by the food industry, these temperatures would be sufficient to sterilise sufficient volumes of sediment to effectively make them barren. Mineralisation within the aureole also increases density, converts sulphate and carbon substrates to less reactive solid phases and decreases porosity. Basin modelling approaches, with improved temperature constraints and improved measures of thermal maturity, would be needed to reliably apply this information to greater volumes of sediment to forecast habitability.

A new method for measurement of geological core sample quality using X-ray Computed Tomography data and its application of IODP Expedition 370

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The evaluation of the quality of the drilled cores is vital for scientific drilling to determine the locality of sampling intervals, to interpret data and consider coring-methods. We propose a new method for a core quality evaluation using X-ray computed tomography data. The method provides quantitative core quality factor (CQF) and Mean CT number (MCN) profile of geological core samples. The CQF is effective to assure the quality of core samples and has some benefits responding to requests for core quality evaluations, for example, the high spatial resolution, quick measurement, and the flexible criteria for evaluations. The MCN profile considering the CQF is reliable, high quality and useful to understand the downhole variation of the major material of core samples. XCT data of core samples from International Ocean Discovery Program (IODP) Site C0023 located at a toe of accretionary complex of the Nankai Trough was treated by this method. The CQF evaluation of cores was a good fit to visual core description and the MCNs of mud/mudstone showed a positive correlation with bulk density data. The MCN profile correspond well visual descriptions and showed several characteristics which reflect to lithology and deformation structures. The distribution of abrupt changes of MCN, are probably due to fault displacements, and show two fault zones concentrated in the Trench to Basin and Lower Shikoku Basin Facies, and also in the upper part the lower part of Lower Shikoku Basin Facies at C0023.

19. Impact of temperature and life on mineralogy (4 x 15 minutes)

- Myriam Kars: Effects of high temperature and a deep SMT on rock magnetic properties
- Natsumi Okutsu
- Kiho Yang: Application of EELS on the microbial mineral alteration
- Oliver Helten & Florence Schubotz: Abiotic substrate generation from recalcitrant organic matter at elevated temperatures

Investigating the effects of high temperature and a deep SMT on rock magnetic properties at Site C0023, IODP Expedition 370

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The aim of IODP Expedition 370 was to explore the limits of life in deep subseafloor sediments in a high temperature environment (up to 120Â°C), and to investigate, among other objectives, the processes at the biotic-abiotic transition. A deep (inverse) sulfate-methane transition (SMT) was identified between 630 and 750 mbsf. Here, a rock magnetic study is conducted to document the downcore variations in magnetic properties and mineralogy (contents, grain size and composition of the magnetic mineral assemblage) related to post-depositional diagenetic processes from 200 to 1100 mbsf, with a focus on the deep SMT. 225 discrete cube samples were collected in the Kochi Core Center (KCC) for this study. Natural remanent magnetization and its alternating-field demagnetization, magnetic susceptibility and acquisition of isothermal remanent magnetization were measured on the samples. Hysteresis properties and first order reversal curves were measured on respective dry powders for magnetic grain size study and composition of the magnetic assemblage. Based on magnetic data profiles and results from previous ODP expeditions in the area, four magnetic zones (Zones 1 to 4) were defined mostly reflecting alteration and diagenetic features. Rock magnetic measurements (low resolution FORC diagrams, thermomagnetic measurements) show complex magnetic mineral assemblages with (Ti)- magnetite as the main carrier of the remanence. Hematite is also present, whereas greigite is only found in the ~200-500 mbsf depth interval. Other accessory minerals (not present everywhere) likely include goethite and pyrrhotite. The rock magnetic results will be presented and discussed based on the shipboard and newly acquired geochemical data.

Progress toward understanding the characteristics and sedimentary system of silt-turbidites of IODP Site C0023

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Turbidity currents are the primary agent for transporting sand, silt and clay particles from the shallow to the deep ocean. This process deposits turbidites. A number of events such as earthquakes, floods and storms can generate turbidity currents, therefore understanding the depositional dynamics of turbidity currents helps to interpret modern-ancient sedimentary systems. Paleocurrent analysis can be a useful method to gain information about flow direction, and more broadly the sedimentary system around a study area (e.g. Kanamatsu, 1996). A previous paleocurrent analysis of the Nankai Trough has shown that turbidity currents are deflected from and reflected against trench slopes (Pickering et al., 1982). The core used in this study was obtained from Site 808, which is situated near to Site C0023. However, detailed interpretations of the deeper parts are still limited. This is due to the ongoing difficulty faced in identifying silt-turbidites. Although previous studies have reported characteristics within the surface core or at outcrops (Stow and Shanmugam, 1980), there are few studies of the characteristics of silt-turbidites within drilled-core. To solve these issues, we first focused on the characteristics of silt-turbidites of Site C0023. For this, we mainly used VCD, X-CT image and Mean CT number (MCN). The second focus is to understand the sedimentary system of Site C0023 estimated from paleocurrent analysis using Anisotropy of Magnetic Susceptibility (AMS) measurements. In this presentation, I will mainly talk about the first part: the characteristics of silt-turbidites of Site C0023.

References

Pickering, K.T., Underwood, M.B. and Taira, A., Proceedings of the Ocean Drilling Program, Scientific results, 131, 35-43 (1993)
Stow, D.A.V. and Shanmugam, G., Sedimentary Geology, 25, 23–42 (1980)
Kanamatsu, T., Proceedings of the Ocean Drilling Program, Scientific Results, 149, 335-352 (1996)

Abiotic substrate generation from recalcitrant organic matter at elevated temperatures at IODP Site C0023 in the Nankai Trough, Japan

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IODP Expedition 370 set out to explore the deep subsurface temperature limit of life and its controls in the high heat flow area of the Nankai Trough (Site C0023). Organic matter contents in sediments at Site C0023 are extremely low (avg. 0.3 wt%; Heuer et al., 2017) and become increasingly graphitized with depth and temperature. The presence of methane and C2+ hydrocarbon gases carrying a thermogenic signal, together with abundant acetate and other volatile fatty acids in sediments situated at temperatures >75 °C, indicate that the thermal degradation of organic matter at Site C0023 releases potential substrates that may sustain the indigenous microbial community. The main aims of this study are (i) to perform a detailed characterization of the quality and bioavailability of organic matter at Site C0023 and (ii) to identify potential substrates that may be released at elevated temperatures over time. For this, we will inquire three different sub-fractions of organic matter (OM): (1) water-extractable OM, (2) solvent-extractable OM and (3) kerogen, using gas chromatography coupled to mass spectrometry and various pyrolytic and spectroscopic techniques. Our results will be put into context of cellular concentrations and pore water geochemistry in order to gain a better understanding on the controls that organic matter has on the sustenance of deep hot subsurface microbial life.