

International Ocean Discovery Program

Expedition 370 Scientific Prospectus

T-Limit of the Deep Biosphere off Muroto (T-Limit)

Deciphering factors that constrain the extent of the deep biosphere in a subduction zone

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Abstract

Determining factors that limit the biomass, diversity, and activity of subseafloor microbial communities is one of the major scientific goals to be addressed by scientific ocean drilling. In the International Ocean Discovery Program (IODP) T-Limit Project, we will drill and core at new boreholes in the immediate vicinity of Ocean Drilling Program (ODP) Sites 1173, 1174, and 808 off Cape Muroto, Japan, in the central Nankai Trough, where anomalously high heat flow regimes result in temperatures of 110° to 140°C at the sediment/basement interface. Because of their location in the trench (Site 1173) and landward prototrust zone of the Nankai Trough accretionary prism (Sites 808 and 1174), the sites have different geotectonic and thermal histories that have resulted in contrasting (bio)geochemical modes of hydrocarbon gas production and consumption. Although the upper temperature limit appears well constrained at relatively energy-rich hydrothermal vent systems at just above 120°C, it is unknown in energy-starved sedimentary subseafloor settings but is generally presumed to be lower and thus expected to be covered by our target sites. During the IODP T-Limit Project, we aim to

- Comprehensively study the factors that control biomass, activity, and diversity of microbial communities in a subseafloor environment where temperatures increase from ~30° to ~130°C and thus likely encompasses the biotic–abiotic transition zone; and
- Determine geochemical, geophysical, and hydrogeological characteristics in sediment and the underlying basaltic basement and elucidate if the supply of fluids containing thermogenic and/or geogenic nutrient and energy substrates may support subseafloor microbial communities in the Nankai accretionary complex.

Because of the D/V *Chikyu*'s schedule, these scientific objectives cannot be achieved within a single expedition. During the first T-Limit expedition (370), we will drill and retrieve core samples from sedimentary sections (200–1210 m below seafloor) and basement basalt (1210–1260 m below seafloor) at the prototrust site near ODP Site 1174 and measure temperatures *in situ*.

Schedule for Expedition 370

The operational schedule for International Ocean Discovery Program (IODP) Expedition 370 is derived from original Integrated Ocean Drilling Program drilling Proposal 865-Full (abstract available at <http://www.iodp.org/800>). Following ranking by the IODP Science Evaluation Panel, the expedition was scheduled for the D/V *Chikyu*, operating under contract with the Japanese Implementing Organization, Center for Deep Earth Exploration (CDEX). The *Chikyu* IODP Board has charged the Project Coordination Team (PCT) with formulating a strategy to achieve the overall scientific objectives outlined in Proposal 865-Full. At the time of publication of this *Scientific Prospectus*, the expedition is scheduled to start with a 3 day port call at Shimizu, Japan, on 10 September 2016; depart Shimizu on 13 September; and return to Kochi, Japan, on 10 November 2016. Details of the operational schedule, updates to the drilling schedule, and operational and layout details of the *Chikyu* can be found at <http://www.jamstec.go.jp/chikyu/e/chikyuexp>. The entire expedition comprises a total of 62 days (including 3 days port call and 6 days contingency time) for drilling and coring operations and installation of a temporary temperature observatory, as

described in this *Scientific Prospectus*. Expedition 370 will focus on operations at proposed Site ODP11-74B and will be the first of two scientific drilling expeditions of the IODP T-Limit project, which together aim to achieve the complete set of scientific objectives described in Proposal 865-Full. The second expedition for the remaining part of T-Limit is yet to be scheduled. The two-expedition approach was chosen in order to increase flexibility in the scheduling of ship time.

Introduction

Determining the vertical extent of the habitable zone on Earth and, by inference, the factors that limit life's maximum depths is a central goal pursued by the international marine and continental deep-biosphere community (IODP New Science Plan [NSP], Challenge 6; <https://www.iodp.org/science-plan-for-2013-2023>). Environmental factors such as temperature, pH, salinity, availability of potential metabolic energy, nutrients, fluid movement, and pore space are known to exert controls on habitability, with temperature commonly used as the variable defining the deepest boundary of the deep biosphere in estimates of its size (Whitman et al., 1998; Parkes et al., 2000; Lipp et al., 2008; Heberling et al., 2010; Kallmeyer et al., 2012; Hinrichs and Inagaki, 2012; Parkes et al., 2014). Several IODP expeditions have addressed factors that potentially limit life in the subseafloor. For example, Integrated Ocean Drilling Program Expedition 329 studied the influence of energy limitation on subseafloor communities in the South Pacific Gyre, where low concentrations of sedimentary organic matter and its refractory nature strongly limit microbial life and result in aerobic subseafloor sedimentary ecosystem with low cell densities at 10^3 – 10^4 cells/cm 3 (D'Hondt et al., 2009, 2015; D'Hondt, Inagaki, Alvarez Zarikian, and the Expedition 329 Scientists, 2011; Røy et al., 2012). Integrated Ocean Drilling Program Expeditions 317 (Canterbury Basin) and 337 (Shimokita Coalbed Biosphere) studied microbial communities in extremely deeply buried sediment at depths close to 2 and 2.5 km below seafloor (kmbsf), respectively (Ciobanu et al., 2014; Inagaki et al., 2015). Whereas both expeditions presented strong evidence for the presence of life throughout the entire drilled sediment sequence, Expedition 337 documented an abrupt decrease in microbial cell concentration at subseafloor depths below ~1.5 kmbsf to levels that are drastically lower than predicted by extrapolation of the global regression line (Inagaki et al., 2015; cf. Parkes et al., 2000, 2014). In spite of the great depths, *in situ* temperatures did not exceed 65°C.

The currently known upper temperature limit of life for micro-organisms inhabiting comparatively energy-rich hydrothermal vent environments is from 113° to 122°C (Blöchl et al., 1997; Kashefi and Lovley, 2003; Takai et al., 2008). On the other hand, estimates relevant for the energy-limited marine sedimentary biosphere are derived from studies of petroleum biodegradation, suggesting that sterilization takes place at formation temperatures between 80° and 90°C (Head et al., 2003; Wilhelms et al., 2001). The lower temperature limit in subsurface settings relative to hydrothermal settings is possibly linked to the enhanced requirement of metabolic energy at elevated temperatures for the repair of degraded biomolecules (cf. Head et al., 2003); the potential supply of metabolic energy is low in sedimentary settings that tend to be lean in electron acceptors and/or donors compared to hydrothermal vents, at which reduced fluids are mixed with oxygenated seawater. Experimental studies with surface sediment demonstrate a nonlinear effect of temperature on biogeochemistry and prokaryotes, with rapid changes occurring over small temperature intervals, and reveal that substrate

addition and increase of metabolic energy result in stimulation of microbial activities at elevated temperatures (Roussel et al., 2015). The influence of temperature on microbial communities was one of the central questions during Integrated Ocean Drilling Program Expedition 331 at the Iheya North hydrothermal field in the Okinawa Trough, but this task was severely complicated by extremely high geothermal gradients of 0.6° to 7.0°C/m. Consequently, microbial life seemed to cease within a few tens of meters below seafloor, and strongly condensed temperature zones prevented in-depth examination of critical depth intervals (Takai, Mottl, Nielsen, and the Expedition 331 Scientists, 2011; Yanagawa et al., 2014). To date, the influence of temperature on the amount, activity, and taxonomic composition of deep subseafloor sedimentary biomass has not been rigorously studied through scientific ocean drilling combined with state-of-the-art investigative techniques.

During the IODP T-Limit Project, we will revisit the Muroto Transect in the Nankai Trough off the Kochi Prefecture, Japan, to explore subseafloor life and the conditions conducive to its sustenance in the vicinity of ODP Sites 1173, 1174, and 808 (Figure F1) (Moore et al., 1991; Moore, Taira, Klaus, et al., 2001). In this area of the Nankai Trough, heat flow is exceptionally high and results in temperatures of ~110°–130°C at the sediment/basement interface at ~0.7–1.2 kmbsf (Moore et al., 1991; Moore and Saffer, 2001) (Figure F2). This particular geological setting will enable in-depth examination of the putative temperature-dependent biotic–abiotic transition zone at relatively shallow depth, but with high temperature resolution; the increase of temperature with depth is still gradual enough for the establishment of distinct depth horizons with suitable conditions for psychrophilic (optimal growth temperature range < 20°C), mesophilic (20°–45°C), thermophilic (45°–80°C) and hyperthermophilic (>80°C) microorganisms (Figure F2).

The Nankai Trough subduction zone has been investigated intensively throughout the history of scientific ocean drilling. The available geochemical, geomicrobiological, geological, and geophysical data as well as the success of the coring operations during ODP Legs 131, 190, and 196 suggest that the vicinity of Sites 1173, 1174, and 808 is ideally suited for in-depth examination of environmental factors that control biomass, activity, and diversity of microbial life and ultimately limit the extent of the deep sedimentary biosphere. For Sites 1173 and 1174, depth profiles of microbial cell concentrations were obtained by manual microscopic cell counting during Leg 190 in 2000. Despite the minimal quantification limit (MQL) of ~10⁵ cells/cm³, cell concentrations appeared to drop abruptly to nondetectable levels at sediment depths around 500 and 800 m below seafloor (mbsf) at Sites 1173 and 1174, respectively, where estimated *in situ* temperatures exceed 85°–90°C (Figures F3; Shipboard Scientific Party, 2001a).

Since Leg 190, drastic advances in cell separation and enumeration technologies for sediment core samples (e.g., Kallmeyer, 2011; Morono et al., 2009, 2013; Morono and Inagaki, 2010) have lowered the MQL by a factor of ~500 or more. Likewise, the new capacities offered in the “omics” era for elucidation of taxonomic composition (e.g., Inagaki et al., 2006; Sogin et al., 2006), metabolic activity, and function at single-cell to community levels (e.g., Biddle et al., 2008; Lloyd et al., 2013; Orsi et al., 2013), as well as new molecular-isotopic techniques that link biomass to substrate pools (e.g., Biddle et al., 2006; Morono et al., 2011; Wegener et al., 2012) and central metabolic intermediates to distinct geomicrobiological processes (e.g., Heuer et al., 2006, 2009; Zhuang et al., 2014; Wang et al., 2015) allow entirely new approaches to study microbial life close to the limit of the deep biosphere. Recent advances in analytical capabilities pro-

vide a tool kit for rigorously examining the unique geosphere–biosphere interactions in geothermally heated sediment that involve the release of microbially utilizable substrates by thermal decomposition of refractory kerogens that otherwise appear resistant to microbial consumption. We are now in a position to monitor the compositional effects of abiotic and biotic processes on sedimentary organic matter as well as the products that may serve as substrates for sustaining deep subseafloor microbial ecosystems. Thermodynamic modeling can then reveal the actual amounts of energy available and necessary to maintain microbial metabolism.

Our recent findings for deep subseafloor sediment cored using the *Chikyu*’s riser-drilling system during the Expedition 337 in the northwestern Pacific coast off the Shimokita Peninsula, Japan, underline the relevance of our research question and highlight the advancement of scientific technologies over the past 15 y. Cell concentrations dropped drastically in ~40°–60°C sediment at depths below ~1.5 kmbsf, but indigenous microbial cells were detected with concentrations ranging from <10 to ~10⁴ cells/cm³ (Figure F4; Inagaki et al., 2015). In addition, taxonomic composition of deep microbial communities differed markedly from those in shallower depths, and peak cell concentrations were found in sediment associated with lignite coalbed layers at ~2.0 and 2.4 kmbsf.

Given those previous technological progresses, the T-Limit Project aims to

- Study the factors that control biomass, activity, and diversity of microbial communities in a subseafloor environment where temperatures increase from ~30° to 130°C and thus likely encompasses the biotic–abiotic transition zone, and
- Determine geochemical, geophysical, and hydrogeological characteristics in sediments and the underlying basaltic basement and elucidate if the supply of fluids containing thermogenic and/or geogenic nutrient and energy substrates may support subseafloor microbial communities in the Nankai accretionary complex.

To this end, drilling of two new sites was proposed. Proposed Site ODP11-74B will be located in the protothrust zone of the accretionary prism in close proximity to Sites 1174 and 808. Proposed Site ODP11-73A will be drilled in the incoming, undeformed sediment of Shikoku Basin near Site 1173. Geothermal heating is expected to result in modern temperatures of ~110°–130°C at the sediment/basement interface of both sites, but their geotectonic and thermal histories are distinctly different (Figure F2; Shipboard Scientific Party, 2001c). The investigation and comparison of both sites will allow us to understand the chemical and physical characteristics of the biotic–abiotic transition zone and to elucidate how temperature affects biogenic and abiogenic processes in the deep subseafloor biosphere over time.

The operational plan of the T-Limit Project is guided by three major objectives:

1. Recovery of high-quality cores from critical temperature intervals of the sedimentary sequence will be accompanied by monitoring of drilling-induced contamination and quality assurance/quality control (QA/QC) during laboratory procedures. High sample quality and careful contamination controls are prerequisites for investigating the presence (or absence) of life at the lower boundary of the deep biosphere.
2. Precise and accurate knowledge of *in situ* temperatures is not only essential for delineating the impact of temperature on subseafloor life but also for characterizing the heat and fluid flow

regime within the Nankai Trough subduction zone. Therefore, in situ temperatures will be determined by downhole measurements during drilling as well as by deployment of a temporary temperature observatory (TTO) for long-term (~1–2 y) observation of critical temperature intervals down to the sediment/basement interface.

3. Recovery of basement core samples will allow us to investigate heat and fluid flow in the subducting oceanic crust and thus the potential introduction of basement-derived microbes, nutrients, and electron acceptors into the overlying sediment column.

In order to fully exploit the technological potential available to date, the scientific work program onboard the *Chikyu* will be complemented by simultaneous analytical work at the Kochi Core Center (KCC), which is located within reach of a helicopter shuttle for transportation of freshly cored samples. The investigation of microbial communities and processes close to the limits of both subseafloor life and the biosphere will in particular benefit from clean-room facilities, aseptic core sampling techniques, and the state-of-the-art analytical infrastructure at KCC, such as nanoscale secondary ion mass spectrometry (NanoSIMS). Scientists onboard the *Chikyu* and at KCC will process samples, share data, and report results together as one team of expedition scientists according to IODP policies.

Because of the *Chikyu*'s schedule, the scientific objectives of the T-Limit project as approved by IODP's Science Evaluation Panel recommendations on Proposal 865-Full cannot be achieved within a single expedition in the near future. In order to increase flexibility, we have split the complete work program into work packages that we aim to accomplish with two expeditions as ship time becomes available during suitable seasons. Expedition 370 is the first expedition of the T-Limit project and will drill one of two proposed sites (ODP11-74B) down to the basement in order to

- Retrieve high-quality sediment and basalt core samples from critical temperature intervals,
- Conduct in situ temperature measurements during drilling, and
- Deploy a TTO into the borehole.

Background

Geologic setting

Located east of Japan between Shikoku Basin and the Southwest Japan arc, the Nankai Trough marks the subduction boundary between the young, hot Philippine Sea plate and the Eurasian plate (Figure F1). Subduction of Shikoku Basin occurs at a current rate of ~2–4 cm/y (Seno et al., 1993). With a maximum water depth of 4900 m, the Nankai Trough is relatively shallow, mainly because a thick sediment pile consisting of turbidite layers on top of hemipelagic basin sediment accumulated on the young (~16 Ma) basaltic basement (Taira, Hill, Firth, et al., 1991; Moore, Taira, Klaus, et al., 2001; Mikada, Becker, Moore, Klaus, et al., 2002). Because of the high flux of terrigenous sediment from the arc to the trench, the Nankai Trough has formed a thick, clastic-dominated accretionary prism over the last ~5 my (Shipboard Scientific Party, 2001a). As the accreted materials are lithified by physical and geochemical processes, the prism accumulates strain energy that is eventually released by seismic activity with a frequency of about one great earthquake every 180 y during historic times (Ando, 1975).

The Nankai Trough has been investigated intensively throughout the history of scientific ocean drilling. Deep Sea Drilling Project (DSDP) Legs 31 and 87 were drilled off Cape Ashizuri to better un-

derstand initial mountain building processes (Figure F1) (Shipboard Scientific Party, 1975; Coulbourn, 1986). ODP Legs 131, 190, and 196 continued these studies with a stronger emphasis on the role of dewatering and fluid flow in the consolidation and deformation of sediment off Cape Muroto where the fault and décollement zone at <1000 mbsf are within reach of riserless drilling technology. Six sites were established along the Muroto Transect (Figure F1), covering undeformed to highly deformed zones of the accretionary prism (Taira, Hill, Firth, et al., 1991; Moore, Taira, Klaus, et al., 2001; Mikada, Becker, Moore, Klaus, et al., 2002). During the past decade, the Nankai Trough Seismogenic Zone Experiment (NanTroSEIZE) has investigated this subduction zone with a multidisciplinary and multiphase drilling, sampling, and monitoring program to better understand the plate boundary fault systems that has historically produced massive earthquakes. About 200 km north of the Muroto Transect, NanTroSEIZE established 13 drilling sites along the Kumano Transect off the Kii Peninsula where the plate boundary fault system is within reach of riser drilling technology. In the course of numerous seismic and bathymetric surveys, the structure of the Nankai Trough off Shikoku was imaged extensively (e.g., Aoki et al., 1982; Ashi and Taira, 1992; Costa Pisani et al., 2005; Karig, 1986; Kodaira et al., 2000; Le Pichon et al., 1987; Moore et al., 1990, 1991; Okino and Kato, 1995; Park et al., 1999, 2000; Stoffa et al., 1992; Taira and Ashi, 1993). A large volume of 3-D seismic reflection data was acquired in 1999 (R/V *Ewing* Cruise 9907/8) in an effort to image the décollement from the trench into the seismogenic zone (e.g., Bangs et al., 1999, 2004, 2005; Gulick et al., 2004).

The Muroto Transect is more suitable for our research objectives than the Kumano Transect, as the deeply buried sediment is exposed to higher temperatures. Located in the trench outer margin, Site 1173 represents a reference site for the incoming, undeformed sediment of Shikoku Basin. Site 1174 is located in the protothrust zone of the accretionary prism (Figure F5). At both sites, ~16 my old basaltic basement is covered with the following series of sediment: volcaniclastic facies, lower Shikoku Basin facies (hemipelagic mudstone), upper Shikoku Basin facies (hemipelagic mudstone with abundant volcanic ash layers), basin-to-trench transition facies, and outer trench wedge facies. At Site 1174, the ~300 m of axial trench wedge facies and slope apron facies have been deposited on top of the outer trench wedge facies (Shipboard Scientific Party, 2001b, 2001c). Both sites subsided at a moderate rate of ~50 m/my during the Miocene and Pliocene, before rates changed dramatically in the Quaternary. Although the rate of subsidence increased to ~75–100 m/my at Site 1173, it rose 12-fold to ~600 m/my at Site 1174 and resulted in a high heating rate of up to 130 K/my (Horsfield et al., 2006). At Site 1173, deformation structures are sparse, but at Site 1174 deformation bands have developed below 218 mbsf, and at ~808–840 mbsf a distinct 32 m thick décollement zone separates the protothrust domain with fractured and steepened bedding (<807.6 mbsf) from a relatively little-deformed underthrust section (>840.2 mbsf) (Shipboard Scientific Party, 2001c).

Heat and fluid flow

There is compelling evidence for hydrothermal circulation in the subducting oceanic crust, which leads to elevated heat flux at the deformation front. Specifically, thermal and geochemical models suggest that advective heat and fluid flow in the subducting oceanic crust are crucial for extracting and redistributing heat (Harris et al., 2013; Spinelli and Wang, 2008), whereas the accreted sediment expels fluids mainly by upward diffusive flow through a large

portion of the accretionary prism (Taira et al., 1991, 1992). Pore water profiles did not clearly indicate active fluid flow along the décollement or along the frontal thrust, and fractures within the décollement zone have not been mineralized (Taira et al., 1991; Maltman et al., 1992; Moore, Taira, Klaus, et al., 2001); however, at >400–500 mbsf, pore water freshening is detectable in Cl^- concentrations that are lower than seawater values (Figures F6, F7; cf. Kastner et al., 1993). The observed freshening is partly explained by in situ diagenesis (compaction and thermally driven smectite dehydration) but also suggests some episodic lateral fluid flow along one or more sediment horizons (Moore, Taira, Klaus, et al., 2001; Saffer and Bekins, 1998; Underwood et al., 1993).

Sites 1173 and 1174 are both located in a zone of high heat flow that encompasses the trench and the lowermost part of the prism (Figure F1). The surface heat flux consistently exceeds 100 mW/m^2 in this region and amounts to ~180 mW/m^2 at Sites 1173 and 1174 but steeply declines landward of the deformation front to values of ~60 mW/m^2 and scatters largely around 82 mW/m^2 seaward in Shikoku Basin (Yamano et al., 1984; Kinoshita and Yamano, 1986; Shipboard Scientific Party, 1991; Shipboard Scientific Party, 2001b, 2001c; Spinelli and Underwood, 2005; Harris et al., 2013; Yamano et al., 1992). In situ temperatures at the sediment/basement interface are expected to be ~110°C at Site 1173 (734 mbsf) and ~120–140°C at Site 1174 (1194 mbsf) based on downhole temperature measurements in the upper 300 m of these sites (Figure F2; Shipboard Scientific Party, 2001b, 2001c).

Physical properties

Measurements of physical properties in sediment and basement rock, particularly their variations with depth, are a major target in studies of rock mechanics and fault zone seismogenesis. With increasing temperature and pressure, physico-chemical interactions are facilitated and cause lithification and diagenesis (Figure F6) (Moore and Saffer, 2001). These processes modify the composition of the fault gouge and result in authigenic growth of clay minerals (e.g., Chester et al., 1993; Wintsch et al., 1995), which are known to have profound effects on fault strength and potential instability (e.g., Byerlee and Summers, 1975; Ikari et al., 2009). In particular, the smectite-to-illite reaction, which is typically observed at 60°–150°C (i.e., temperatures typical of the up-dip seismicity limit [Vrolijk, 1990]), releases fluids and silica into the pore space. Released silica acts as a cementing agent (Towe, 1962; Spinelli et al., 2007) and thus strengthens the frictional properties of the materials (e.g., Mair and Marone, 1999) while lowering their effective porosity (Spinelli et al., 2007). At the same time, the release of mineral-derived water from desorption and clay mineral transformation reactions into an environment in which precipitation has decreased porosity may cause high pore pressure and potential hydrofracturing of the rock (Behrmann, 1991). Elevated pressures and temperatures typical of upper seismogenic zones are also known to enhance porosity loss in sediment (Hüpers and Kopf, 2009) and are associated with decreases in permeability. The result is a dense, cemented rock that differs substantially from homogeneous sediment and may thus impose additional limits on the deep biosphere.

Microbiology and geochemistry

Whereas previous investigations focused on the dynamics of deformation and fluid flow processes in the accretionary prism, the abundance of microbial cells in sediment was measured manually by microscopy on board the R/V *JOIDES Resolution* (Figure F3), and the geochemistry of sediment and its interstitial water were

characterized as well (Moore, Taira, Klaus, et al., 2001). Cell concentrations were somewhat higher at Site 1173 (Figure F7) than at Site 1174 (Figure F8). At Site 1174, the first drop of cell counts below the MQL was observed at ~550 mbsf, which corresponds to a temperature of ~65°C. Interestingly, counts exceeded the MQL again in deeper sediment just above the décollement around 800 mbsf where temperatures reached 90°C; no cell detection was reported below that depth. Cell concentrations at Site 1173 were in a range typical for continental margin sediment downhole to ~500 mbsf (~85°C) but dropped to nondetectable levels below that depth.

At both sites, organic matter contents decrease with depth. In particular, the transition from upper to lower Shikoku Basin facies corresponds to a drop in total organic carbon (TOC) content. Carbon and nitrogen ratios <10 indicate predominantly marine organic matter sources. Pore water sulfate is consumed rapidly in surface sediment and a shallow sulfate–methane transition zone (SMTZ) is found at 3–6 mbsf, but sulfate concentrations increase again throughout the ~490 m thick lower Shikoku Basin sediment and linear concentration gradients are consistent with diffusive flux of sulfate from the oceanic basement into the overlying sediment (Shipboard Scientific Party, 2001b, 2001c; Horsfield et al., 2006).

The two sites differ distinctly with respect to the biogenic and/or thermogenic alteration of organic matter during burial. At Site 1173, methane concentrations are high in the upper 100 mbsf at temperatures around 18°C but decrease to lower concentrations downhole to 300 mbsf and are very low deeper than 350 mbsf, where methane is possibly consumed as a result of sulfate diffusion from the basement (Figure F7) (Shipboard Scientific Party, 2001b). In contrast, methane concentrations are highest at 300–660 mbsf at Site 1174 (~60°–100°C) and remain relatively high in the presence of sulfate below 660 mbsf (Figure F8) (Shipboard Scientific Party, 2001c). At Site 1173, high methane concentrations go along with peak values of alkalinity, ammonium, and phosphate concentrations, suggesting together that organic matter remineralization is most intense in the upper 100 mbsf. In contrast, the deeper methane maximum at Site 1174 is decoupled from alkalinity and ammonium concentrations, which peak within the upper 300 mbsf. At both sites, ethane concentrations increase with depth and point to thermogenic sources below 350 and 900 mbsf at Site 1173 and 1174, respectively (Shipboard Scientific Party, 2001b, 2001c; Horsfield et al., 2006). This observation agrees well with findings at the adjacent Site 808, where the stable carbon and hydrogen isotopic composition of methane points to biogenic CO_2 reduction as the major methane source in sediment between 20 and 1000 mbsf and to an increasing contribution of thermogenic methanogenesis at greater depth (Berner and Faber, 1993). At Site 1173, concentrations of molecular hydrogen and acetate, both potential substrates for microorganisms, increase deeper than 350 mbsf (Parkes et al., 2007). Results of Rock-Eval pyrolysis reveal significant thermal transformation of organic matter below 300 mbsf at Site 1173 and below 500 mbsf at Site 1174, suggesting the production of hydrocarbon gases, hydrogen, and oxygenated low-molecular weight compounds in situ (Horsfield et al., 2006). Yet, the interplay of thermogenic and biogenic processes and their relevance for the supply of metabolic energy remain to be explored.

Overarching scientific objectives and hypotheses

The scientific objectives in the T-Limit project are highly relevant to Challenge 6 in the IODP New Science Plan

(<https://www.iodp.org/science-plan-for-2013-2023>) “What are the limits of life in the subseafloor?” and relevant to Challenge 5 “What are the origin, composition, and global significance of subseafloor communities?”

The overarching scientific objectives are as follows.

1. Study subseafloor sedimentary microbial communities situated in temperature ranges that cover the putative temperature limit of microbial life in anoxic sedimentary systems.

We hypothesize that temperature increase with depth will be accompanied by substantial changes in the composition, function, and activity of microbial communities and that their population density decreases gradually rather than abruptly as suggested by previous examination of these sites. Today, the state-of-the-art cell enumeration assays and molecular ecological and geochemical techniques will provide a far more comprehensive view of the relationship between microbial communities and temperature than was possible during Leg 190.

2. Characterize the chemical and physical environment in sediment and basement rock at the limit of the deep biosphere.

We hypothesize that the sections to be drilled harbor biotic–abiotic transition zones and that we can identify unique geochemical and microbial signatures within the sedimentary, rock, and fluid matrixes that differentiate the biotic and abiotic realms and/or their transition. In addition to the techniques routinely employed across the entire drilled section, sediment crossing the putative biotic–abiotic transition zone will be investigated by a combination of techniques that will enable a detailed examination of the (bio)chemical, isotopic, mineralogical, and physical properties and changes thereof.

3. Examine the relationship between thermogenic release of potential substrates and microbial life.

We will test existing hypotheses (Horsfield et al., 2006; Parkes et al., 2007) that predict stimulation of microbial activity by thermal decomposition of organic matter. To that end, we will conduct studies of genes, their expression, cultivation-dependent and -independent assays, biogeochemical activity measurements, and advanced organic geochemical analyses that specifically target potential substrates for microbial life, as well as document structural modifications of macromolecular organic matter induced by their release. Comparison of proposed Sites ODP11-74B and ODP11-73A will allow us to investigate

- a. Whether their different geotectonic and thermal histories are reflected in microbial community composition,
- b. How the transition from predominantly biogenic (Site 1173) to predominantly thermogenic (Site 1174) gas formation is related to the early thermal alteration of kerogen, and
- c. How the potential stimulation or inhibition of microbes by thermal alteration of organic matter changes over time.

Expedition 370 scientific objectives and questions

Expedition 370, as the first expedition of the T-Limit project, will specifically address the following questions:

- Do subseafloor sedimentary microbial communities populate sediments that lie within the temperature range close to the putative limit of microbial life?

- Is the temperature increase with depth accompanied by substantial changes in the composition, function, and activity of microbial communities?
- Does the population density of microbial communities decrease gradually or abruptly with increasing temperature? And do features like porosity or fluid movement and the introduction of electron acceptors influence the location of the limit to life such that the biotic fringe “flickers out” over a certain depth range rather than “blinks out abruptly” at a specific depth? For example, is microbial activity stimulated by fluid flow along the décollement?
- Are ubiquitous thermophilic spores (cf. Hubert et al., 2009) abundant, and do they germinate when temperatures increase with sediment burial?
- Which microorganisms and processes occur in the deep SMTZ at ~600–700 mbsf (~75°C)?

Characterizing the chemical and physical environment in sediment and basement rock at the biotic fringe will allow us to address questions such as

- Which mechanisms guide the selection of microorganisms at elevated temperatures?
- Are there other adaptations evident in cells present at the biotic fringe besides tolerance to high temperatures?
- Do pairs of electron acceptors and donors accumulate that would normally be consumed by biologically mediated redox reactions?
- Is there a measurable impact of smectite–illite transition on subseafloor microbial communities?
- Does the presumed availability of electron acceptor-rich seawater in the basement stimulate microbial activity in the overlying sediment? If there are active (or survived) microbial communities in the high-temperature basaltic habitat, is there evidence for the inoculation of overlying sediment with basement-derived microbes?
- What is the extent and distribution of hydrothermal veining and crustal alteration related to circulation of seawater? How much of this alteration may be related to microbial activity?

Drilling and coring operations

The general operations plan and time estimates are provided in Table **T1** and Figure **F9**. Although site survey data analysis, contingency planning, engineering, and working out the technical details for placing instruments in the boreholes are ongoing at the time of writing this *Scientific Prospectus*, the prioritization of the main activities and overall sequencing is not expected to change.

The main operations to be completed during Expedition 370 are to drill, core, and install a TTO at a single location, proposed Site ODP11-74B. During the expedition, selected core samples will be transported to KCC via helicopter. The sequencing of operations reflects the prioritization of the activities and engineering constraints. The sequence of operations consist of the following:

1. Drilling and running casing through the uppermost 140 m.
2. Continuous coring with hydraulic piston coring system (HPCS) or extended shoe coring system (ESCS) from ~200 mbsf to the top of basement basalt (~1210 mbsf). The modified HPCS will also be used at key intervals (see below). If possible, formation temperature measurement with a probe at regular intervals of 100 m.

3. Setting casing to 1210 mbsf.
4. Continuing coring ~50 m into the basement with the rotary core barrel (RCB).
5. Running tubing and completion assembly (including in situ temperature sensor loggers with the autonomous string) to 1210 mbsf.

Coring plan

Coring starts at 200 mbsf, after the first casing is set to 140 mbsf. Coring with HPCS or ESCS continues through the sedimentary section to the top of basement basalt at ~1210 mbsf. Coring operations continue for another 50 m into the basement. It will be necessary to switch to RCB either once reaching the basement or once the formation is too hard for ESCS.

To study the limit of the deep biosphere, it is crucial to collect contamination-free, high-quality core samples from brecciated fault zone sediment, which typically are difficult to recover in good condition. In case the quality of ESCS cores does not meet the scientific requirements, use of the short-stroke HPCS may be attempted at key intervals. The short-stroke HPCS is still under modification. Technical and operational details are unavailable as of this writing. However, the successful deployment of a similar system, the half-length advanced piston corer, by the *JOIDES Resolution* in recent expeditions (e.g., Expedition 351 Scientists, 2015) suggests that the modified HPCS would be a promising option.

Observatory plan

The installation plan and technical details of the TTO are not yet finalized. The following is the most likely scenario. After coring operation is completed, the 4.5 inch diameter casing will be extended to the top of basement (~1210 mbsf). A string will be constructed by attaching multiple temperature sensor loggers to a Vectran rope. The string of ~50 sensors will have a TTO-hanger attachment on the top and will be fixed to the wellhead for later retrieval (Figure F10). The sensors will be spaced following lithologic and structural changes through the borehole. The exact length of the temperature string and location of the sensors will be decided on board after looking at the results of coring. To retrieve the data, the sensor string be recovered by either the *Chikyu* or other remotely operated vehicle-deployed research vessel (e.g., R/V *Kaiko*).

If budget allows, the 4.5 inch tubing will be composed of an outside string of several thermistors with data telemetered to a data logger at the wellhead. The battery life of the data loggers is expected to be ~5 y. This system will allow data retrieval without physical recovery of the thermistor string as long as the string remains functional. A robust cable will be used so that if there is a break in any portion of the cable, data will still be recorded for portions above the break. The exact depth of the sensors will be decided on board before deployment.

Temperature measurements

To meet scientific objectives for the temperature limit of subseafloor life, measurement of in situ temperature will provide the primary geophysical information of the microbial habitat, but it is also significant to characterize fluid flow regimes that possibly sustain habitability in high-temperature environments. In addition, drilling down to the basement in this region, with high relevance to the prediction of geodynamic processes such as gigantic earthquakes and tsunamis, will provide useful borehole information, especially at

proposed Site ODP11-74B, for potential future missions regarding long-term monitoring connected to the Dense Oceanfloor Network System for Earthquakes and Tsunamis 2 (DONET-2).

During Expedition 370, we plan to measure in situ temperatures using the advanced piston corer temperature tool (APCT-3) and, if possible, with a sediment temperature probe in parallel with coring operations, in addition to long-term monitoring with TTO (and thermistors).

Analytical research plans

To identify unique signatures for the existence and absence of life, we will collect essential data that allow us to assess how close to or far from equilibrium various deep horizons are so that we can inventory the fluid and energy supply and correlate it with molecular information on indigenous microbial communities. Since Leg 190 in 2000, analytical techniques and sensitivity have advanced tremendously, particularly in molecular microbiology, cell enumeration, and molecular and isotopic geochemical techniques. In addition to the standard core descriptions, the general approaches and techniques intended to address the expedition objectives are summarized (but not limited to) as below.

Cultivation-independent microbiological approaches

Detection and enumeration of microbial cells, spores, and activity markers and determination of the composition and function of microbial communities will benefit from vast recent improvements in cell detection and quantification and leaps in various “omics” and chemical (e.g., membrane lipid analysis) approaches that can decipher the composition, function, and activity of complex natural communities. Using newly developed cell detachment and computer-based enumeration techniques, cell concentrations in marine sediment can be precisely determined as low as ~100 cells/cm³ or less instead of ~50,000 cells/cm³ during Leg 190. Novel less-biased DNA extraction, purification, and quantification methods have been established (e.g., Hoshino and Inagaki, 2012; Morono et al., 2014; Lever et al., 2015), and subsequent molecular ecological analyses such as amplicon sequencing (to determine community composition), shotgun metagenomic analyses (to determine potential community metabolisms), metatranscriptomics (to identify gene expression), and metaproteomics (to identify active metabolism and functions for a microbial community) provide a means to explore in great detail shifts in microbial communities at single cells to community levels (e.g., Biddle et al., 2008; Lloyd et al., 2013; Orsi et al., 2013) and potential evolutionary changes for related taxa in a lithostratigraphic context.

Cultivation-based microbiological approaches

Cultivation-based experiments will be designed to isolate microbes, examine indigenous spores, determine process rates and substrate turnover times using radiotracer-based assays, and track the flow of selected substrates into specific biomolecules as well as single cells using stable isotope and radioisotope probing assays on NanoSIMS, together with experiments to constrain the role of biology in kerogen-to-substrate degradation processes, as well as the associated compositional changes of the organic matter pool, under the broad range of in situ temperatures encountered at the two target sites.

Organic matter, potential substrates, and metabolites

In the targeted system, the principal sources of reactants that can provide metabolic energy through microbially mediated redox reactions are provided by the thermal and biological degradation of macromolecular organic matter, both of which result in a broad spectrum of defined and undefined lower molecular weight molecules. We seek to track key degradation processes and structural modifications starting at the kerogen level down to small molecules that are well-defined substrates for microbial metabolism such as acetic acid. Nuclear magnetic resonance spectroscopy analyses will be used to track changes in kerogen chemistry and differentiate between thermogenic and biogenic modes of kerogen decomposition coupled to methane formation (Kamga et al., 2014), whereas ultra-high-resolution mass spectrometry will be used to identify changes in the population of dissolved organic molecules. Isotope-ratio-monitoring mass spectrometry will serve to study the distribution and isotopic composition of low molecular weight organic compounds to identify manifestations of microbial activity or the absence thereof.

Electron acceptors, nutrients, and major anions and cations

Knowing the concentrations of various electron acceptors and donors is a prerequisite for a systematic understanding of the energetic constraints for microbial life in the deep biosphere. We will generate concentration data for redox-relevant dissolved species such as sulfate, nitrate (particularly when approaching basement), metals, dissolved inorganic and organic carbon, selected small molecules such as acetate, formate, carbon monoxide, hydrogen, sulfide, ammonium, methane, and other hydrocarbon gases, and the nutrient phosphate. To a large degree, we will take advantage of established shipboard procedures (e.g., Expedition 329 Scientists, 2011; Expedition 337 Scientists, 2013). Although the concentrations are useful for interpreting data summarized above, the concentration patterns can also directly contribute toward Objective 2 in that we expect the putative biotic–abiotic transition zone(s) to be marked by the absence of microbial turnover of these compounds. Objective 2 will additionally be addressed by the examination of isotopic composition of sulfur species and phosphate, including multiple-sulfur-isotope approaches and dual-isotope analysis of sulfur and oxygen in sulfate and oxygen in phosphate. Recent studies (Ono et al., 2012; Rudnicki et al., 2001; Sim et al. 2011) suggest that multiple-sulfur-isotope measurements are a powerful tool to trace the extent and temperature of microbial sulfate reduction. Likewise, oxygen isotope ratios of sulfate and phosphate will provide critical temperature information because oxygen isotope exchange is very slow below $\sim 250^{\circ}\text{C}$; therefore, at low temperatures equilibrium-like fractionations for $^{18}\text{O}/^{16}\text{O}$ ratios are only achieved in nature by microbial-catalyzed isotope exchange (e.g., Blake et al., 2001; Wortmann et al., 2007; Goldhammer et al., 2011; Brunner et al., 2012). Collectively, these approaches will aid identification of biosignatures, or the absence thereof, in critical sediment horizons around the putative biotic fringe.

Mineral biosignatures

We will employ state-of-the-art mineralogical examinations to identify diagnostic signatures of microbial metabolism on the surfaces of sediment particles in sediment intervals presumably harboring the biotic fringe. These analyses are part of our strategy to

address Objective 2 and complement other approaches that, if isolated because of intrinsic limitations such as detection limits, may not provide an unambiguous picture regarding the presence or absence of life. The general concept of mineral-based biosignatures is described, for example, by Boston et al. (2001). We propose to study prepared samples with a combination of analytical techniques consisting of but not limited to vertical scanning interferometry with a coupled Raman probe (laser power will be delivered at the sample surface well below the critical dose of radiation that can damage the carbonaceous matter); fast-scanning atomic force microscope; scanning electron microscope equipped with secondary-electron, backscattered, and cathodoluminescence detectors as well as a powerful energy dispersive X-ray analyzer system; and electron microprobe. The combination of these techniques will provide both 3-D topographical information and chemical measurements and will enable identification of surface corrosion and/or elemental compositional anomalies caused by microbial life.

Thermodynamic modeling

Chemical energy is available in an environment if there are compounds that are out of equilibrium with one another and can be released if the compounds are allowed to react. By catalyzing oxidation-reduction reactions that are far from equilibrium yet slow to react on their own, chemotrophic microbes can harvest the energy required for their metabolism. Whether a reaction is capable of supplying energy, as well as the quantity of energy available, can be evaluated by thermodynamic calculations at the temperatures and pressures of subsurface conditions. These calculations require analytical data for compounds that could be combined in oxidation-reduction reactions and equilibrium constants for the same reactions calculated independently with standard state thermodynamic data. A comparison reveals the amounts of energy available to fuel microbial metabolism and how quantities of energy change with temperature, pressure, and differences in composition (McCollom and Shock, 1997; Amend and Shock, 2001; Amend et al., 2003; Shock et al., 2010).

QA/QC for microbiological samples

We will implement a rigorous program for contamination monitoring combined with minimization of potential contamination of precious samples. This is a key prerequisite for deep biosphere research in very deep boreholes into consolidated formation and will directly influence the mission's success. Fortunately, we can take advantage of protocols developed during previous expeditions (e.g., Inagaki, Hinrichs, Kubo, and the Expedition 337 Scientists, 2013; Inagaki et al., 2015): directly after core retrieval, core sections will be immediately scanned using X-ray computed tomography (X-CT; as available on the *Chikyu*) to select nondisturbed massive core fragments for cell counts and other geochemical and microbiological analyses. Given the importance of ultraclean subsampling, critical samples will be transferred to KCC and sampled using the aseptic superclean room technology. This approach will enable ultrasensitive cell counts and molecular analyses for sediment samples with very low or even no biomass. Potential contamination of samples by drilling will be assessed with methods that rely on the addition of a perfluorocarbon tracer (PFT) to the drilling fluid (Smith et al., 2000; Lever et al., 2006; Masui et al., 2009; Inagaki et al., 2012; Yanagawa et al., 2013), and various negative controls will be collected during shipboard and shore-based sample processing to identify and assess laboratory-derived contamination sources

(e.g., lab air, water, and chemicals) that may interfere with accurate molecular and geochemical analyses.

Sampling and sample coordination

All shipboard sample and data requests will be coordinated by the Sample Allocation Committee (SAC) with the understanding that the limited data and samples available will require T-Limit Project scientists to collaborate closely. Shipboard and shore-based researchers should also refer to the IODP Sample, Data, and Obligations Policy and Implementation Guidelines at <http://www.iodp.org/program-documents/> for additional details about obtaining and using samples.

Sample requests and coordination

The SAC is composed of the expedition Co-Chief Scientists, Expedition Project Manager, the shipboard curatorial representative, and the IODP curators on shore. The SAC for the expedition must approve access to core samples and data requested during the expedition and during the 1 y moratorium, which starts at the end of the drilling expedition.

Science Party

To achieve the scientific objectives of the T-Limit project, Expedition 370 will invite a total of 25–30 scientists who will be assigned to either the shipboard or shore-based team. Both teams will jointly implement the expedition measurement plan (Figure F11).

Shipboard team

The shipboard team will be in charge of sampling, QA/QC including contamination assessments, time-sensitive (bio-)geochemical and microbiological analyses, and IODP standard measurements in the onboard *Chikyu* laboratories. For the shipboard team, Expedition 370 will invite 20–25 scientists, including two of the three Co-Chief Scientists, sedimentologists, organic and inorganic geochemists/biogeochemists, (geo-)microbiologists, physical property specialists, paleomagnetists/biostratigraphers, petrologists, structural geologists, and hydrogeologists (temperature monitoring specialists). In addition to typical requirements in each of these specialties, particular needs in Expedition 370 include, but are not limited to, the following specific expertise:

- Biogeochemists and geochemists with experience/background in pore water analysis of biologically relevant chemical species, in rate measurements using radioactive tracers, in hydrogen and hydrocarbon gas analysis, and in geochemical modeling of transport and reaction of dissolved constituents;
- Microbial ecologists and molecular biologists with experience in shipboard cell enumeration, in shipboard anaerobic and aseptic sampling of sediment and rock core samples, in shipboard contamination assessment using tracers such as PFT, in cultivation or cultivation-based molecular approaches, and/or in single cell biology and (meta)genomics;
- Paleomagnetists and biostratigraphers with experience in age determination and hole correlations in the northeastern Pacific; and
- Temperature monitoring specialists with experience in borehole observatory and in situ temperature measurements.

Shore-based team

The shore-based team will conduct an advanced set of measurements, which requires shore-based facilities at the KCC, on fresh samples directly sent from the *Chikyu*. The team's activity will mainly focus on microbiological and geochemical analyses, but analytical work of other expertise will also be considered if proposed.

For the shore-based team, Expedition 370 will invite 5–10 scientists, including one of three Co-Chief Scientists. Particular needs include, but are not limited to, the following specific expertise:

Geochemists with experience in quantitative and compositional analysis of dissolved organic matter; and

Microbial ecologists and molecular biologists with experience in shore-based microbiological subsampling, anaerobic cultivation techniques (e.g., using high-pressure chamber and flow-through bioreactor), DNA/RNA-based molecular analyses, and/or single cell-biology and omics approaches

The shore-based team's activity will start 2 weeks after the start of the expedition and is expected to continue for the same length of period with the shipboard team. The activity will be closed 2 weeks after the end of expedition, when the team prepares a draft report that will be merged into the IODP Expedition Report.

Sample and data requests

All Expedition 370 scientists, including shipboard and shore-based teams, must submit at least one data or sample request in advance of the drilling expedition. Additional requests may also be submitted before, during, or after the expedition as necessary to acquire samples or data that will help address fundamental questions of Expedition 370 or individual research projects. Sample or data requests may also be submitted by additional shore-based scientists, but in case of overlap or potential conflict, the Expedition 370 scientists will be given top priority. The initial requests provide the basis for the SAC to develop an integrated program to meet all of the essential postexpedition research objectives and to avoid unnecessary duplication of effort. The initial plan, of course, will be subject to modification depending upon the actual material/data recovered and on collaborations that may evolve between scientists before and during the expedition(s). Modifications to this plan during the expedition require the approval of the SAC. To provide time for the SAC to develop a detailed and integrated sampling strategy, data/sample requests are due by 31 August 2016. Shipboard and shore-based scientists should use the Sample/Data Request form (<http://web.iodp.tamu.edu/sdrm>) to submit their requests.

Contingency plans

Unforeseen circumstances could result in insufficient time being available to complete the entire operations plan. Examples include collapse of a borehole because of difficult formation conditions (e.g., unstable sands), hazardous weather, and hardware failures. In anticipation of challenging and fluctuating environmental conditions, we have included 6 days of contingency for the entire expedition in the operations plan and time estimate. In case further delay in operation takes place, we will adjust the time by reducing the number of cores. Details of possible operational priorities or contingencies will be decided by the Co-Chief Scientists, Expedition/Project Manager, and Operations Superintendents according to operational realities on board the ship.

Expedition scientists and scientific participants

The current list of participants for Expedition 370 can be found at <http://www.jamstec.go.jp/chikyu/e/chikyuexp>.

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Table T1. Operations plan, Expedition 370. PDM = positive displacement motor, BHA = bottom-hole assembly, UWTV = underwater television, POOH = pull out of hole, HPCS = hydraulic piston coring system, ESCS = extended shoe coring system, RIH = run in hole, BTM = bottom, CGR/GR = completion guide roller/guide roller, CSG = casing, RCB = rotary core barrel, TBG = tubing, MTL = miniature temperature logger, CHRT = casing hanger running tool, WOW = wait on weather.

Site	Water depth (mbsl)	Operations	Days	Subtotal (days)	Total (days)
ODP11-74B	4750	1. Transit from Shimizu to proposed Site ODP11-74B (250 nmi)	1.50		1.50
		2. Deploy transponders, preparation	1.00		2.50
		3. Set 20 inch conductor casing and reentry cone and housing at 140 mbsf		5.00	7.50
		a. Run 20 inch conductor casing and reentry guide and housing with underreamer and PDM BHA inside casing at low-current area	1.00		
		b. Recover UWTV, set 20 inch conductor casing at 140 mbsf	2.00		
		c. POOH running tool to above seabed	0.50		
		d. Drift to low-current area, POOH to surface, rig up guide horn	1.50		
		4. Drill and cut core to 1210 mbsf (sediment)		22.00	29.50
		a. Run coring assembly (HPCS) during drifting to coring point	1.00		
		Entry into guide with UWTV and recover UWTV prior to coring			
		b. Drill to 200 mbsf with center bit	0.50		
		c. Cut core from 200 to 700 mbsf (HPCS/ESCS)	8.50		
		d. Cut core from 700 to 900 mbsf (ESCS)	4.00		
		e. Cut core from 900 m to 1210 mbsf (ESCS)	6.50		
		f. Spot kill mud, POOH to above seabed	0.50		
		g. POOH to surface during drifting to low-current area	1.00		
		5. Open hole to 17-1/2 inches, set 13-3/8 inch casing		11.50	41.00
		a. Make up and run in hole 17-1/2 inch open hole BHA, reentry and RIH	1.00		
		b. Open hole to 17-1/2 inches to 1210 mbsf	4.50		
		c. Spot kill mud, POOH to above seabed	0.50		
		d. POOH to surface during drifting to low-current area	1.00		
		e. Dismantle guide horn and install CGR/GR	0.75		
		f. Make up and run 13-3/8 inch CSG above seafloor, reentry and RIH to BTM, set hanger	2.00		
		g. POOH setting tool to surface	1.00		
		h. Dismantle CGR/GR and install guide horn	0.75		
		6. Cut core 50 m in the basement		5.25	46.25
		a. Make up and RIH RCB coring system during drifting to coring point	1.00		
		b. Reentry, RIH to 1210 mbsf	0.50		
		c. Cut core from 1210 to 1260 mbsf (RCB)	1.50		
		d. Spot kill mud, POOH to above seabed	0.50		
		e. POOH to surface during drifting to low-current area	1.00		
		f. Dismantle guide horn and install CGR/GR	0.75		
		7. Completion		3.50	49.75
		a. Run TBG, MTL, set up CHRT	1.25		
		b. Run completion assembly with running string	0.75		
		c. Run UWTV, reentry, resume run and set completion assembly	0.75		
		d. Release running tool, wind up UWTV, pull out running string	0.75		
		8. Retrieve transponders, rig down guide horn	1.00		50.75
		9. Transit from the site to Kochi	0.50		51.25
		10. WOW, mechanical downtime		7.00	58.25
		a. Mechanical downtime	2.00		
		b. WOW	5.00		1.9 months

Figure F1. Map showing heat flow data and ODP/IODP transects and sites in the Nankai Trough (modified from Harris et al., 2013). Marine probe (circles), boreholes (stars), and bottom-simulating reflector (BSR) (small circles) are color coded by heat flow. On land, circles show borehole values of heat flow.

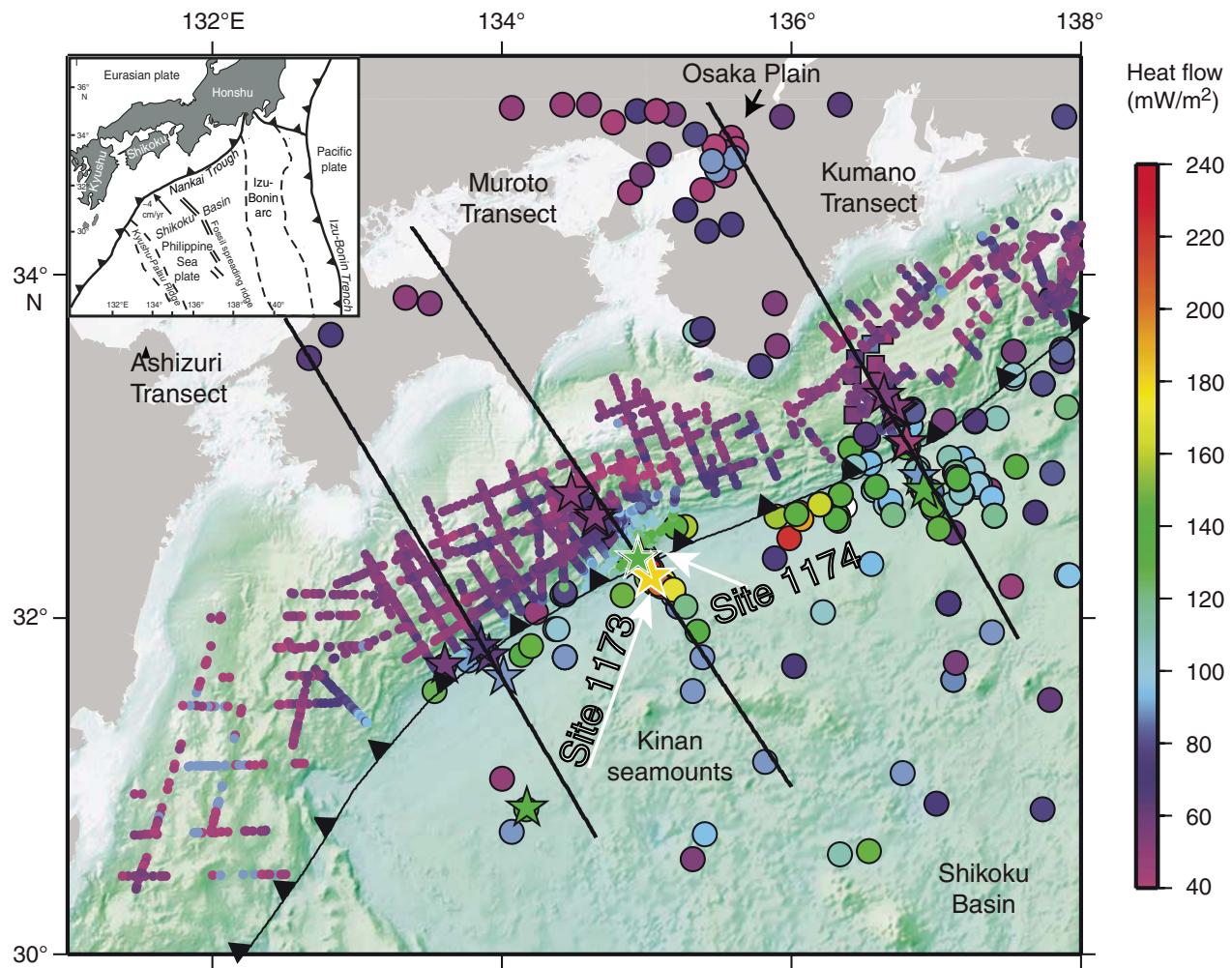


Figure F2. Projected critical temperature window ($T > 80^{\circ}\text{C}$) at Sites 1173 and 1174, based on measured (symbols) and predicted (lines) temperatures (modified from Moore, Taira, Klaus, et al., 2001).

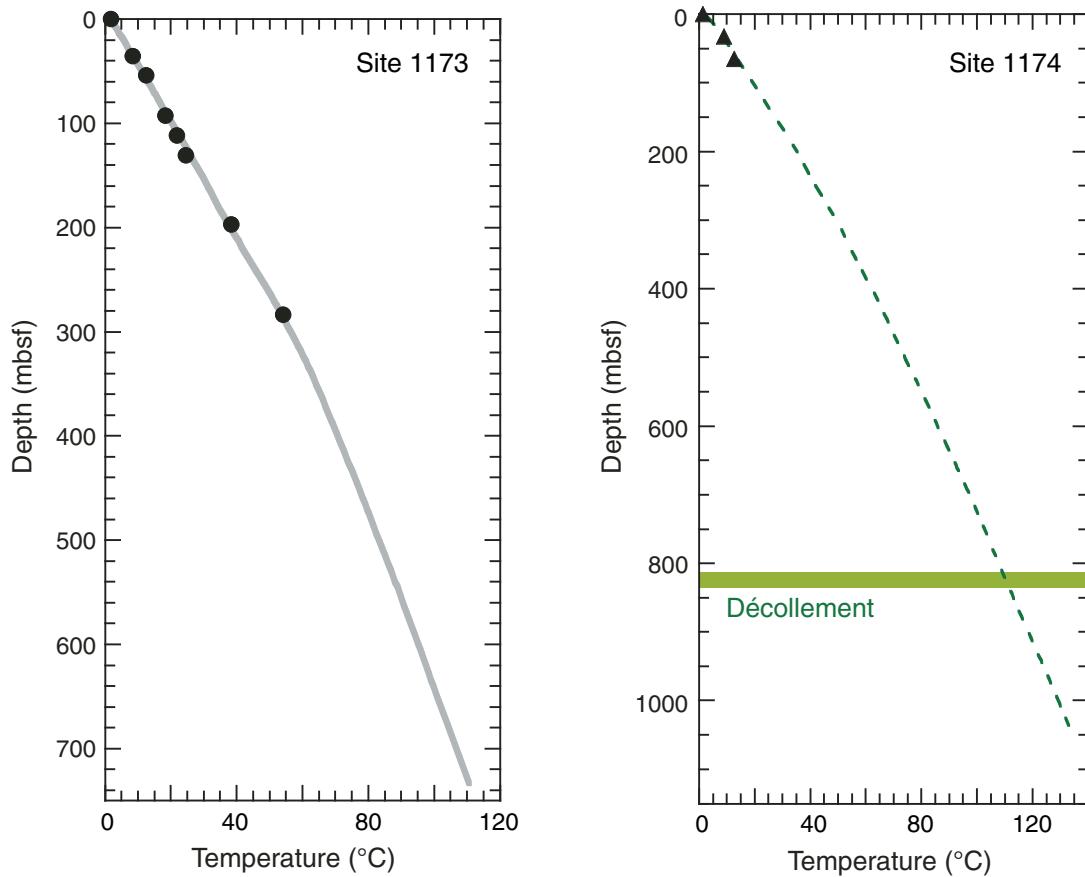


Figure F3. Cell count plots for Sites 1173 and 1174 with depth and temperature axes (Shipboard Scientific Party, 2001b, 2001c). At Site 1174, the first drop of cell counts below the minimum quantification limit (MQL) occurs around 550 mbsf, which corresponds to $\sim 65^{\circ}\text{C}$. Interestingly, counts exceeded the MQL again in deeper sediments just above the décollement around 800 mbsf where temperatures reached 90°C ; no cell detection was reported below that depth. Cell concentrations at Site 1173 were in a range typical for continental margin sediment until ~ 500 mbsf ($\sim 85^{\circ}\text{C}$) but dropped to nondetectable levels below that depth.

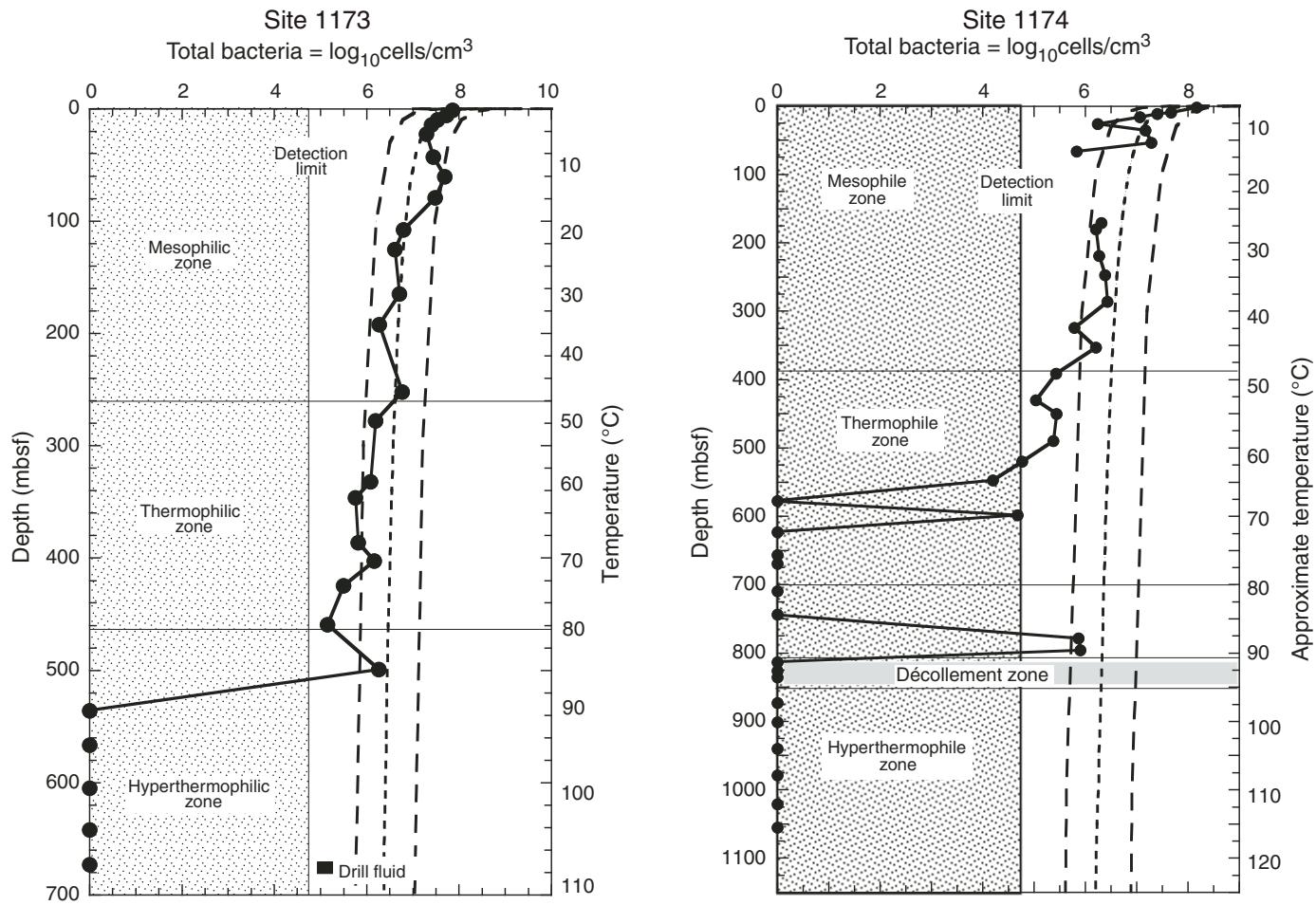


Figure F4. Improvement of cell detection and enumeration technologies for microbial communities in subseafloor sediments. Cell concentration data are derived from previous drilling sites (Parkes et al., 2000, 2014; Kallmeyer et al., 2012; Morono et al., 2009; Inagaki et al., 2015). The minimum quantification limit (MQL) during ODP Leg 190 in 2000 was $\sim 10^5$ cells/cm 3 , which is currently drastically lowered to ≤ 10 cells/cm 3 under the super-clean technology at the Kochi Institute for Core Sample Research, JAMSTEC.

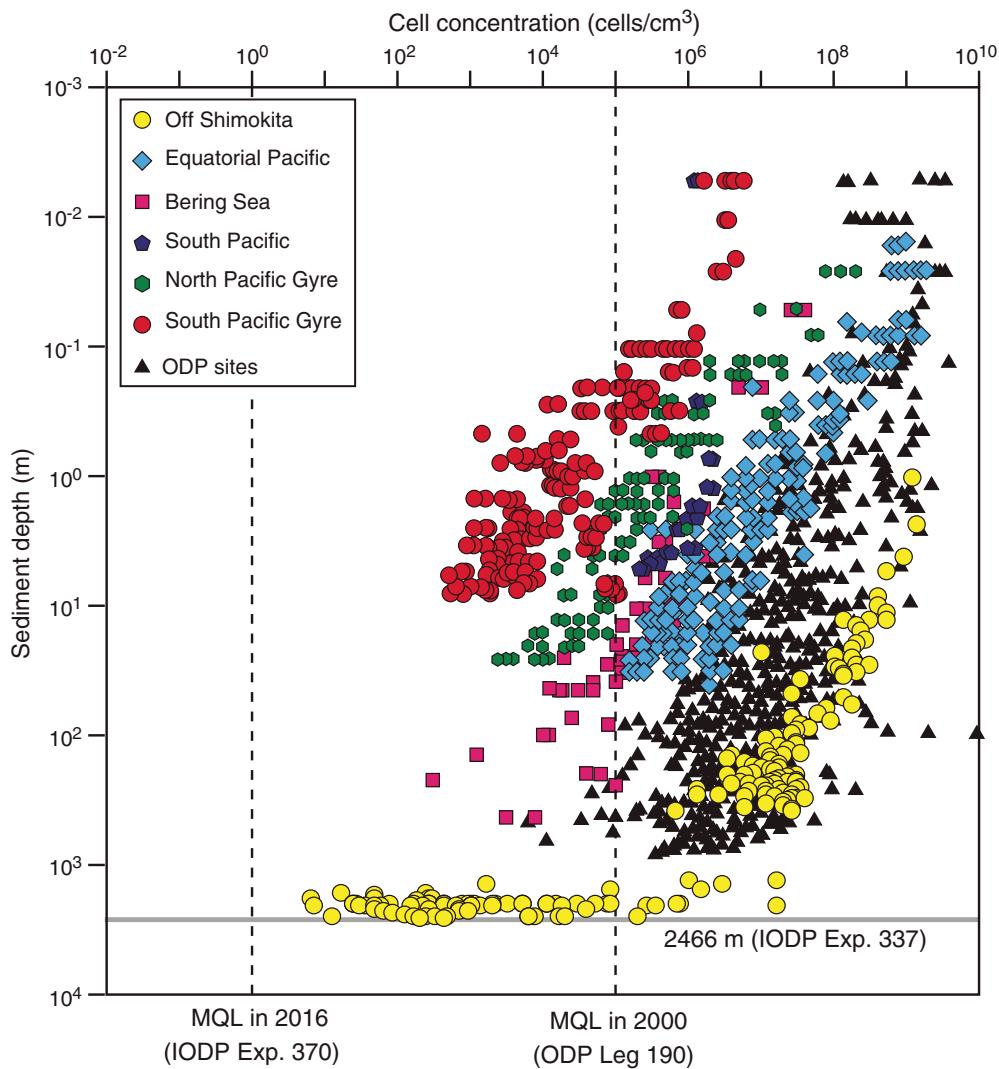


Figure F5. Geological context of the Muroto Transect. Seismic reflection profile through the Muroto Transect reference (ODP Site 1173) and prism toe sites (ODP Sites 1174 and 808). Figure from Shipboard Scientific Party (2001a). Seismic data are from the 3-D seismic survey of Bangs et al. (1999) and Moore et al. (1999). XLine = crossing line number in the 3-D seismic volume.

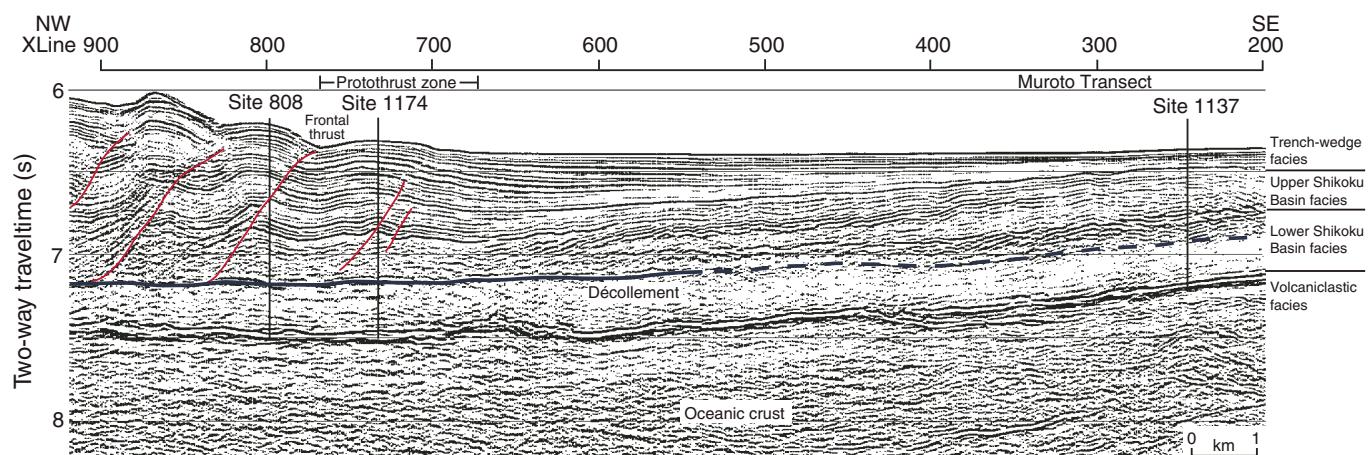


Figure F6. Site 1173: Temperature, lithology, cell counts and geochemical profiles (lithostratigraphy plot and data from Shipboard Scientific Party, 2001b).

Site 1173

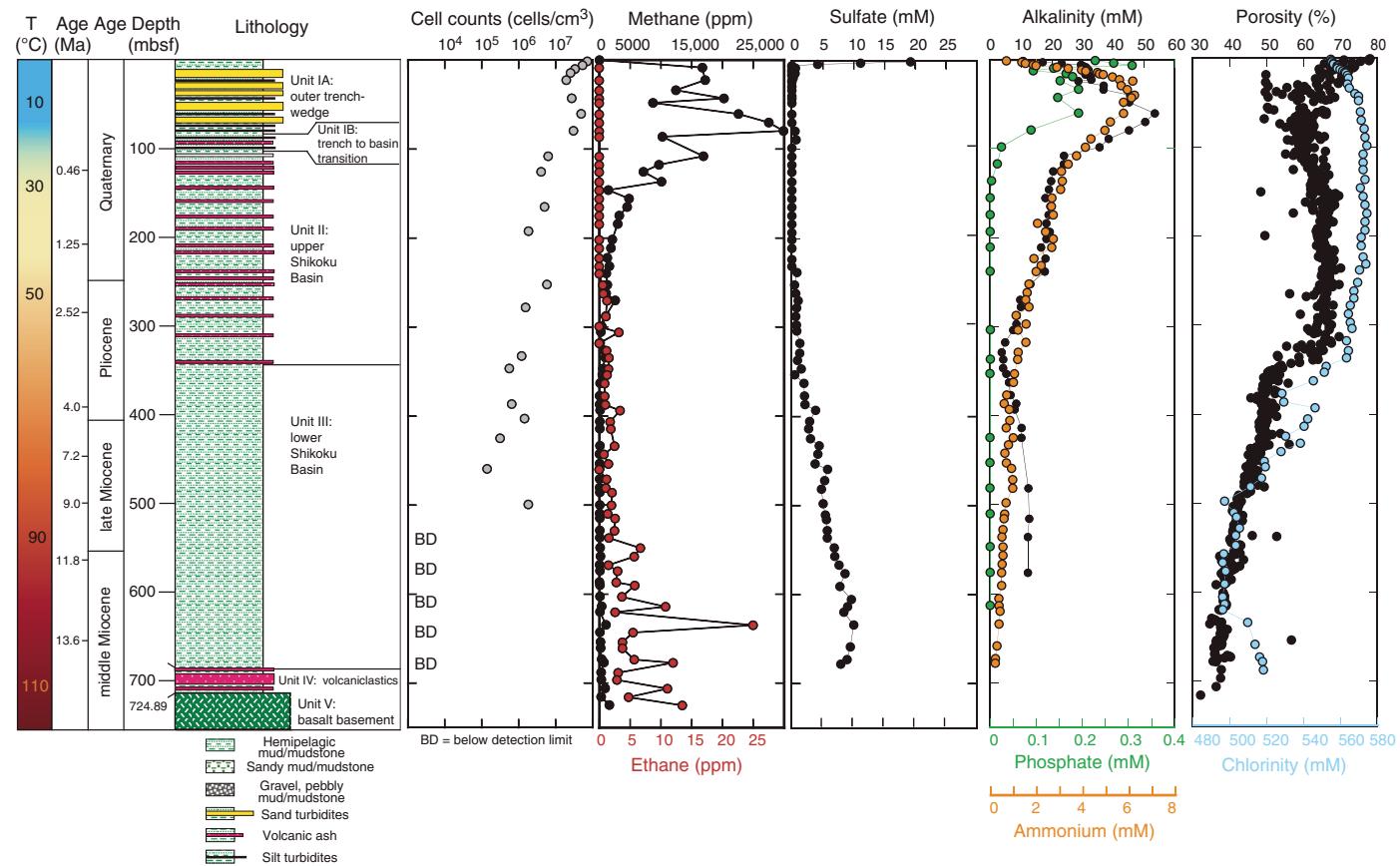


Figure F7. Site 1174: Temperature, lithology, cell counts and geochemical profiles (lithostratigraphy plot and data from Shipboard Scientific Party, 2001c).

Site 1174

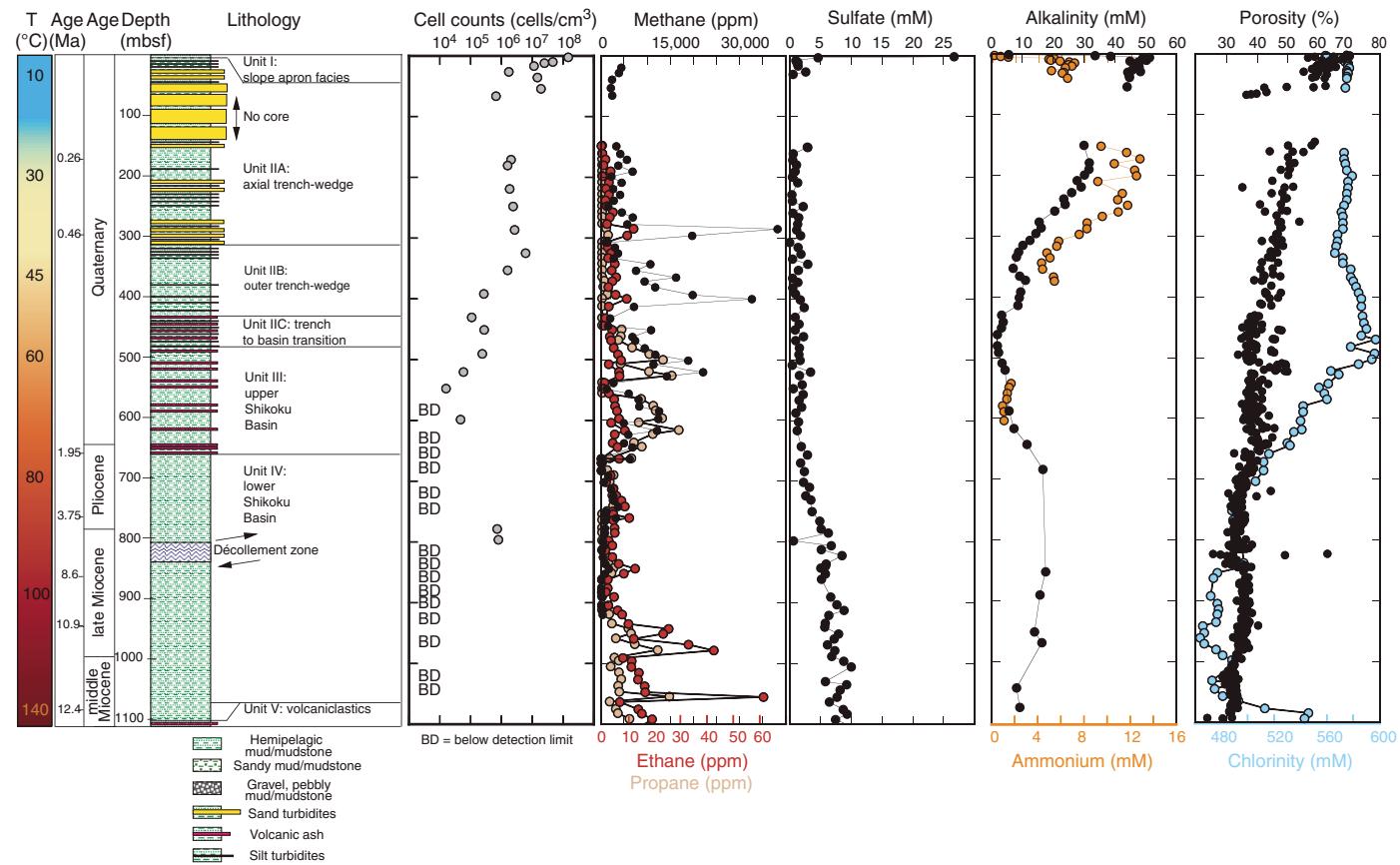


Figure F8. Diagenetic and low-grade metamorphic processes operating in the upper few kilometers of the Earth crust. The shaded area marks the expected critical temperature range at Sites 1173 and 1174. Modified from Moore and Saffer (2001).

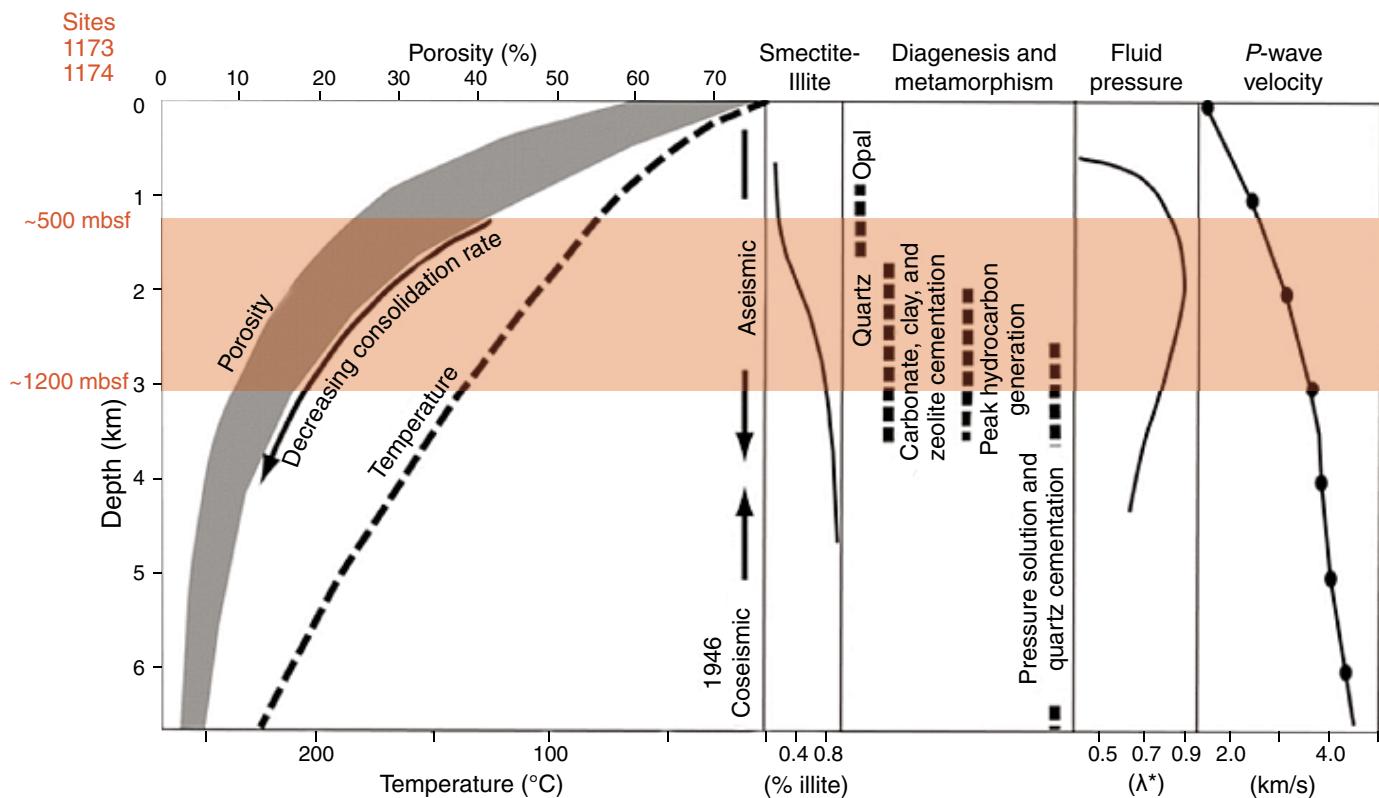
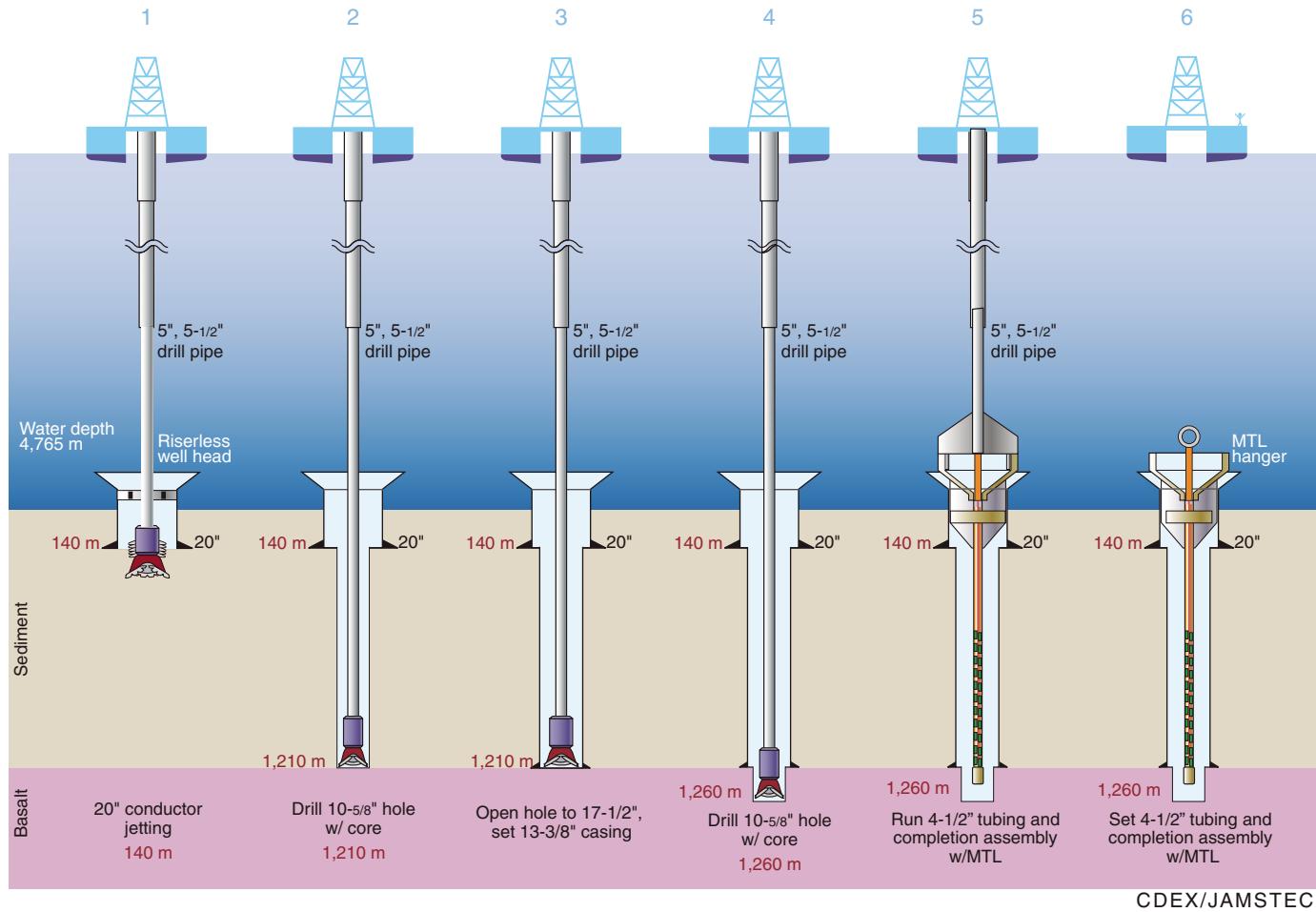


Figure F9. General sequence of Expedition 370 drilling operation.



CDEX/JAMSTEC

Figure F10. Diagram of casing hanger wellhead at the top of the temporary temperature observatory (TTO) with the removable hanger and hanger ring from which the TTO sensor string hangs. Remotely operated vehicle (ROV) will be able to grasp the hanger ring to remove the sensor string and recover the data recorded within each temperature logger. Design and figure are modified from those used during Expedition 343 (Expedition 343/343T Scientists, 2013). MTL = miniature temperature logger.

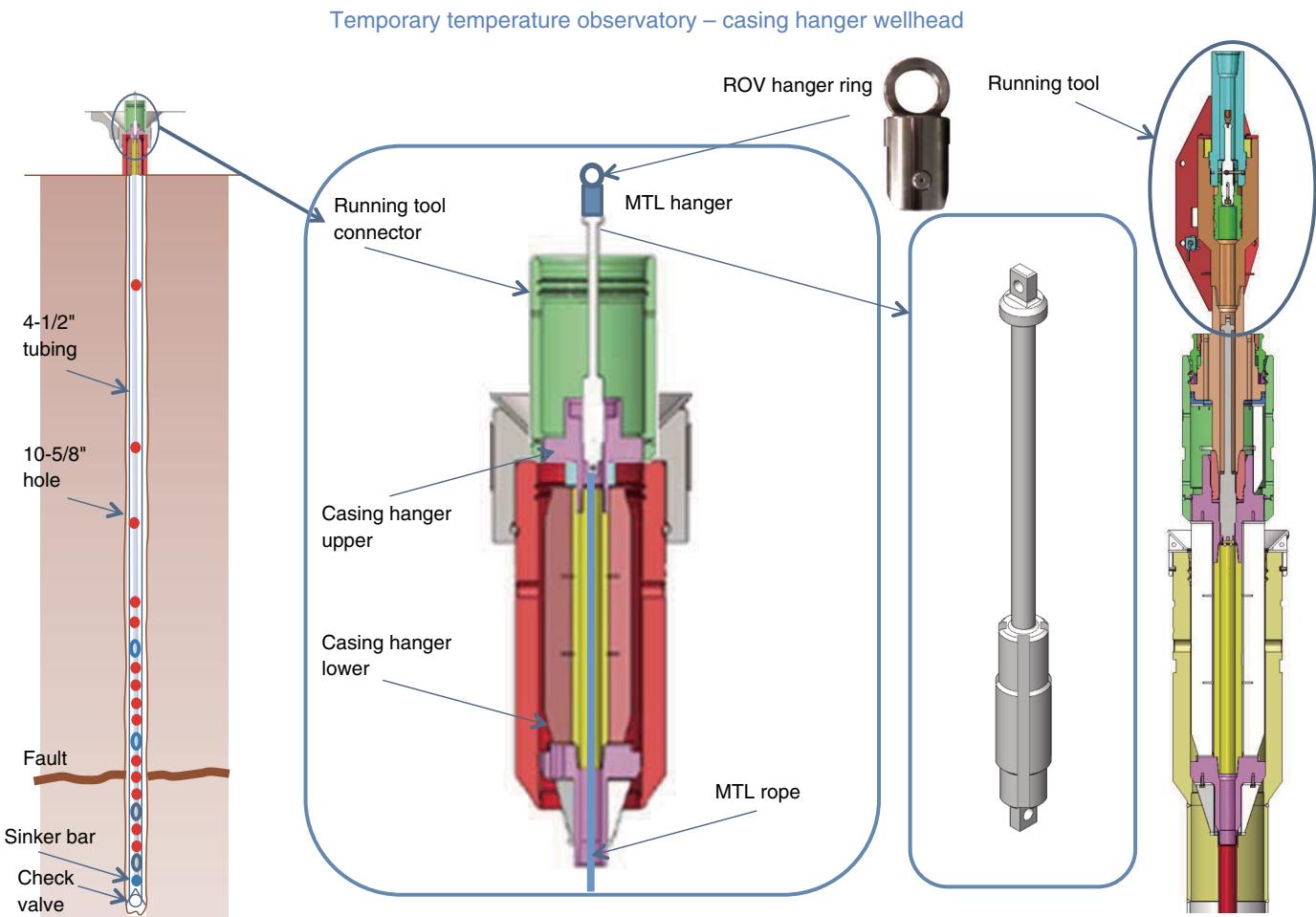
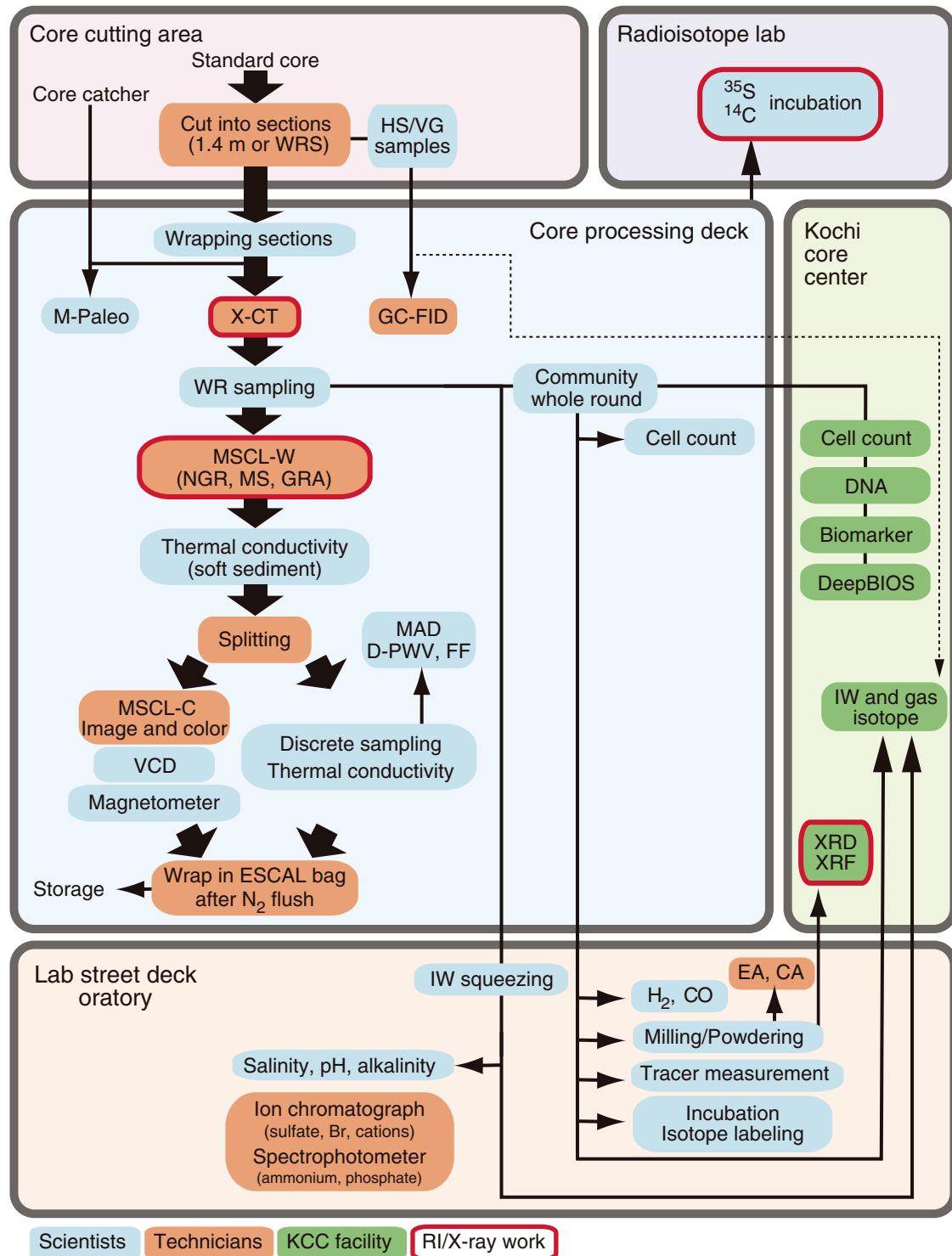


Figure F11. Core analysis flow planned for use during Expedition 370. Note that part of the analysis will be carried out at the shore-based facility at Kochi Core Center (KCC). WRS = whole-round section, HS/VG = headspace/void gas, X-CT = X-ray computed tomography, GC-FID = gas chromatograph–flame ionization detector, WR = whole round, MSCL-W = whole-round multisensor core logger, NGR = natural gamma radiation, MS = magnetic susceptibility, GRA = gamma ray attenuation, MAD = moisture and density, D-PWV = discrete P-wave velocity, FF = formation factor, MSCL-C = color spectroscopy multisensor core logger, VCD = visual core description, DeepBIOS = deep biosphere samples, IW = interstitial water, XRD = X-ray diffraction, XRF = X-ray fluorescence, EA = elemental analysis, CA = carbonate analysis.

Expedition 370 measurement plan



Site summary

Site ODP11-74B

Priority:	Primary
Position:	32°22'00.5678"N 134°57'59.5804"E
Water depth (m):	4765
Target drilling depth (mbsf):	1260
Survey coverage (track map; seismic profile):	Extensive survey data from 3-D seismic data: <ul style="list-style-type: none"> • In-line 332 • Cross-line 781
Objectives:	<ul style="list-style-type: none"> • To study the factors that control biomass, activity and diversity of microbial communities • To determine geochemical, geophysical, and hydrogeological characteristics in sediment and the underlying basaltic basement
Drilling program:	Hole A: install 20 inch casing to ~140 mbsf; coring from 200 to 1210 mbsf; install 13½ inch casing; coring to 1260 mbsf; install temperature monitoring string
Nature of rock anticipated:	Hemipelagic mud/mudstone with turbidite and volcanic ash, highly fractured in part; décollement zone at 870–900 mbsf; sediment/basalt interface at 1210 mbsf