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| **Macintosh HD:Users:Rosalynn:Desktop:C-DEBIlogo4_EEE.eps** | **CENTER FOR DARK ENERGY BIOSPHERE INVESTIGATIONS  STC Annual Report 2016** |

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# I. GENERAL INFORMATION

## 1. Center General Information

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| --- | --- |
| **Date Submitted** | 12/31/2016 |
| **Reporting Period** | 1/1/2016 - 12/31/2016 |
| **Center** | Center for Dark Energy Biosphere Investigations |
| **Center Director** | Jan P. Amend |
| **Lead University** | University of Southern California |
| **Co-Principal Investigator** | Steven L. D’Hondt, University of Rhode Island |
| **Co-Principal Investigator** | Andrew T. Fisher, University of California Santa Cruz |
| **Co-Principal Investigator** | Julie A. Huber, Marine Biological Laboratory |
| **Co-Principal Investigator** | C. Geoffrey Wheat, University of Alaska Fairbanks |

## 2. Changes in Faculty

None.

## 3. Primary Contact

|  |  |
| --- | --- |
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## 4. Context Statement Statement

The mission of C-DEBI is to explore life beneath the seafloor and make transformative discoveries that advance science, benefit society, and inspire people of all ages and origins. Specifically, we seek to better understand the organisms that inhabit the sediment, rock, and fluid in the marine subsurface. Our scientific goals are pursued with a combination of approaches, through which we: (1) coordinate, integrate, support, and extend the research associated with major field programs, including at the Juan de Fuca Ridge flank (JdF), South Pacific Gyre (SPG), North Pond (NP), Dorado Outcrop (DO), and the Atlantis Massif (AM); (2) make substantial investments of resources to support field, laboratory, analytical, and modeling studies of the deep subseafloor ecosystems; (3) facilitate and encourage synthesis and thematic understanding of submarine microbiological processes, through funding of scientific and technical activities, coordination and hosting of meetings and workshops, and support of researchers and graduate students; and (4) entrain, educate, inspire, and mentor an interdisciplinary community of researchers and educators, with an emphasis on undergraduate and graduate students and early-career scientists. In our education and outreach projects, we create and operate programs aimed at three target groups: (1) undergraduate students, especially community college students, (2) graduate students and postdoctoral scholars, and (3) K-12 and the general public.

In 2016, significant accomplishments were recorded for the five major field programs, as well as for other field, laboratory, and modeling projects. For example, new findings that span hydrogeology, biogeochemistry, and microbial ecology at JdF were reported, including the properties and processes that control the dynamics of a multi-outcrop (tens of kilometers apart) hydrothermal siphon capable of transporting heat and solutes within the upper ocean crust; the first cross-hole tracer experiment (using a dissolved gas tracer) in the volcanic ocean crust; and several investigations assessing the microbial community, their *in situ* activities, and the bioenergetics landscape in JdF crustal fluids.

The SPG program produced several scientific accomplishments this year, published in peer-reviewed journals. For example, we demonstrated experimentally that H2 production per unit of radiation is amplified by up to a factor of 33 in abyssal clay, making radiolytic H2 an especially important electron donor for microbes in organic-poor sediment. We showed that bacterial diversity in anoxic subseafloor sediment declines exponentially with sediment age, in parallel with total community respiration and energy flux. We concluded that some marine microfossil DNA survives for more than a million years, and that radiolytic H2 may be the primary electron donor in fractures of SPG basalt older than ten million years. Lastly, we published contamination-tracing results for IODP Expedition 329, providing necessary context for all studies of the SPG microbial ecosystem.

Research at NP in 2016 continued to focus on sediment, rock, and fluid samples from the 2011 drilling expedition and the 2012 and 2014 ROV programs. We showed that the microbial communities in the oxygenated crustal aquifer at NP are distinct from those in the overlying sediment and from those in bottom seawater. It was demonstrated that nitrogen can stimulate the growth of basalt-associated microbes, and three aerobic heterotrophs were isolated and partially characterized from NP sediment samples. A laboratory study further validated aerobic respiration in the crustal fluids at NP.

The relatively new investigation at DO has started to generate new and exciting findings about the subseafloor. One submitted manuscript highlights the spring fluid composition, revealing that discharge from cool ridge-flank systems can source substantial fluxes of Rb, Mo, V, U, Mg, Li and other metals on a global scale. The first microbial ecology study published species richness and diversity data of mineral-attached communities. It was noted that the diversity, dominated by chemolithoautotrophs, is greater than that observed in the surrounding seawater. A third paper elucidates the regional subsurface hydrologic flow, building on a three-dimensional model developed with data from the JdF system. Lastly, a handful of other papers are in preparation, ranging from hydrogeologic investigations to organic and inorganic fluid analyses to sediment subsurface microbial ecology studies.

The offshore phase of the AM expedition concluded only a year ago, and the onshore phase followed in early 2016. This expedition was the first ever to successfully use *seabed* drilling, recovering tens of meters of core from 17 holes drilled at 9 sites across the massif. Initial data show that wide-scale serpentinization is on-going and that H2-rich fluids likely exist in the basement rocks. Microbial activity experiments, initiated in the offshore phase, are targeting a variety of biogeochemical cycles, with a particular focus on methane and sulfur cycling.

While laboratory growth studies of deep marine subsurface isolates are only just beginning, biogeochemical modeling efforts have produced a number of published papers and manuscripts. In several investigations on a global scale, we demonstrated that marine sediments contribute significantly to global element cycles on multiple time scales; this is due in large part to microbial activity in the shallower layers and to abiotic reactions as temperatures and pressures increase with ever-deeper burial. We also quantified the amount of energy required by microorganisms to make biomass as a function of temperature, redox state, and identity and concentration of C, N, and S sources. Lastly, we described microbial degradation of organic matter in marine sediments on multiple time scales, suggesting that there are major uncertainties regarding the rates at which microbes process organic carbon with depth and time on a global scale.

As noted, C-DEBI is also directly involved in several other field-based studies (e.g., Baltic Sea Basin; off Cape Muroto, Japan; Mariana serpentinite mud volcanism; Mariana Back-Arc), and C-DEBI supports numerous projects through a grants and fellowships program. This program funds small research studies, short-term personnel exchanges, education and outreach activities, and fellowships. In 2016, 36 such projects were active, including 12 new research awards made in 2016 that focus on a variety of topics, including the diversity of extracellular electron transfer in marine sediment, the ability of hydroxyl radicals to liberate bioavailable organic carbon in sediments, the identities and single-cell activity rates of diazotrophic microbes in sediments, the biogeochemistry of fluids from the serpentinite subsurface, and the energy yields of 1000 potential metabolisms within the compositional variation of oceanic crusts near mid-ocean ridges.

C-DEBI’s mailing list counts approximately 1000 individuals in nearly 30 countries. Of these, over 250 are 'active' participants, defined as those who presented or participated at one of our workshops or conference sessions, submitted proposals to C-DEBI, served on one of our committees or panels, or otherwise engaged directly in C-DEBI science or education activities. Our focus on junior researchers is demonstrated by the 16 graduate and postdoctoral fellowships that were active during this reporting period. Our commitment to growing research and education on life beneath the seafloor is demonstrated by the fact that 45 individuals representing 26 institutions received funding this year through the C-DEBI grants and fellowships program.

C-DEBI’s education, outreach, and diversity (EOD) programs focused predominantly on research opportunities for community college students, and training and mentoring of graduate and postdoctoral researchers. Two of our EOD flagship programs are the Community College Research Internship for Scientific Engagement (CC-RISE) and the Community College Cultivation Cohort (C4). CC-RISE is a non-residential REU-style program at two of our partner institutions, the University of California at Santa Cruz (UCSC) and the Marine Biological Laboratory (MBL) in Woods Hole, Massachusetts. Twelve students spent 8 weeks during the summer in state-of-the-art research labs, carrying out experiments and analyses, and participating in professional development seminars. C4 is a residential REU program where 8 students spent 9 weeks during the summer characterizing the physiology and phylogeny of 3 novel subseafloor bacterial isolates, learning culturing, microscopy, analytical chemistry, and bioinformatics in the process. As noted above, C-DEBI supports a number of graduate students and postdoctoral scholars through our fellowship program. In addition, these groups of young scientists make up the bulk of the research personnel in the Co-I and Senior Scientists labs. In 2016, several dozen graduate students and postdoctoral scholars worked on C-DEBI projects in the Amend, Huber, Fisher, Wheat, D’Hondt, Orphan, Orcutt, Spormann, Finkel, and Heidelberg labs. They received a wide range of professional training that intertwined their research with education and outreach.

As C-DEBI transitioned to Phase 2, we incorporated a number of changes. The leadership group expanded to add more strength in microbiology, and the research agenda achieved a balance of field-based, laboratory, and modeling investigations. We also honed our EOD programs in undergraduate, graduate, and postdoctoral activities, and we integrated these activities more directly with our research mission. Further, our knowledge transfer (KT) and data management and integration (DMI) teams have more clearly defined goals and objectives, with a particular focus on archiving, sharing, and making publically available in a timely fashion all C-DEBI data and products. In this regard, we have established a close partnership with the Biological and Chemical Oceanography Data Management Office (BCO-DMO). Lastly, we have replaced most members of our external advisory board (EAB)—only Susan Humphris as Chair has remained—and in the process focused the expertise in microbial ecology research, community college education, and private foundation funding opportunities.

# II. RESEARCH

## 1. Overall Research Goals and Objectives

C-DEBI’s central research goal is to investigate the marine deep subsurface biosphere.  While we’ve made substantial progress over the past few years, much remains unknown about the physiology, phylogeny, distribution, limits, and activity in the sediments, rocks, and fluids that make up this very large biome.  C-DEBI continues to generate knowledge in this area by addressing key questions, which include:

* What are the nature and extent of life in the subseafloor?
* What are the physico-chemical limits of life in the subseafloor?
* How metabolically active is the subseafloor biosphere?
* What are the dominant redox processes in the subseafloor?

In Phase 1 (2010-2015) of C-DEBI, our research efforts focused predominantly on four major field programs at the Juan de Fuca Ridge flank, the ocean floor below the South Pacific Gyre, a sedimented site in the North Atlantic called North Pond, and the Dorado basaltic outcrop in the eastern equatorial Pacific. As we transitioned into Phase 2 (2015-2020), we diversified our research portfolio to include other field sites, including, but not limited to a targeted study at the ocean core complex of the Atlantis Massif in the North Atlantic. In Phase 2, we are also complementing our field-based research with laboratory studies of microbial survival and propagation, and modeling studies of physico-chemical properties in the subsurface.

To achieve C-DEBI objectives, we directed a large portion of the research funds to the Co-Investigators (Jan Amend, USC; Julie Huber, MBL; Steven D’Hondt, URI; Andrew Fisher, UCSC; Geoff Wheat, UAF) and Senior Scientists (Steven Finkel and John Heidelberg, USC; Beth Orcutt, Bigelow; Victoria Orphan, Caltech; Alfred Spormann, Stanford); substantial resources also went to competitively awarded research grants, and graduate student and postdoctoral fellowships.

In Phase 2, our research framework balances discovery science, where the activities are dominated by field measurements, instrument development and deployment, and sample analysis, with hypothesis testing, data integration, laboratory experimentation, and ecosystem modeling. The overarching research themes and associated objectives require highly multidisciplinary and interdisciplinary approaches, with the greatest emphasis on microbial ecology. The research themes are:

***Theme 1: Fluxes, Connectivity, and Energy***—centering on subseafloor environmental conditions.

(1.1) Constrain the extent, variability, and controls on fluxes and connectivity within subseafloor biomes and between the subseafloor and the overlying ocean.

(1.2) Map the geochemical energy sources in subseafloor ecosystems at a range of spatial scales.

(1.3) Develop and test the next generation of coupled geochemical-hydrological-microbial models for subseafloor ecosystems.

***Theme 2: Activities, Communities, and Ecosystems***—emphasizing resident microbial communities.

(2.1) Determine community composition, functional potential, and patterns of natural selection in subseafloor ecosystems.

(2.2) Determine metabolic activity of subseafloor microbial communities.

(2.3) Advance understanding of subseafloor microbe-virus interactions.

***Theme 3: Metabolism, Survival, and Adaptation***—concentrating on the actions and traits of individual

microbial species.

(3.1) Isolate and characterize novel bacteria and archaea from diverse subseafloor habitats.

(3.2) Examine fundamental physiology of subseafloor microbes under conditions of low growth rates and low energy flux.

(3.3) Perform adaptive evolution and long-term survival experiments with subseafloor microbes to characterize molecular genetic signatures associated with particular phenotypes.

**2. Research Thrust Areas**

Here, we summarize the most important research accomplishments in 2016; we encountered no noteworthy problems. The first five subsections (2.a-e) cover major field programs at the Juan de Fuca Ridge Flank (JdF), South Pacific Gyre (SPG), North Pond (NP), Dorado Outcrop (DO), and Atlantis Massif (AM). We provide a brief background on each site and describe the key operational, scientific, and technical accomplishments. The next three subsections focus on other field projects (2.f), important laboratory studies (2.g), and modeling (2.h). The last two subsections briefly highlight projects funded through our grants and fellowship program (2.i) and workshops supported by C-DEBI (2.j).

**a. Field Site: Eastern Flank of the Juan de Fuca Ridge**

**Led by:** Andrew Fisher, University of California at Santa Cruz

**Background**

The Juan de Fuca Ridge flank (JdF) major program is exploring the nature of linked hydrogeologic, geochemical, and microbiological conditions and processes within a region of young, volcanic ocean crust that is mostly buried by thick and continuous sediments. This is one of the best-studied ridge-flank systems on Earth, and provides particular value when compared to other ridge-flank sites at NP and DO. These two systems are cool and oxic, whereas the JdF is warm (up to ~65°C) and highly reacted (suboxic to anoxic). Although sediment restricts the movement of fluids, heat, and solutes between the volcanic crust and ocean throughout most of the JdF region, isolated volcanic outcrops (seamounts) penetrate the sediment in a few locations, providing focused flow pathways. Experiments on the JdF were a primary focus in Phase 1 (following Expedition 327), and longer term studies were organized through use of sealed, subseafloor observatory systems (CORKs) that were visited after drilling for several years as part of submersible expeditions. Materials and data collected during the drilling expedition and subsequent CORK servicing expeditions have been used for laboratory analyses, experiments, and modeling. In addition, tools and methods developed for the JdF program were adapted and applied at other sites, including numerous C-DEBI program locations. The JdF major program has also contributed to training of numerous graduate and undergraduate students and postdoctoral researchers, including those who add diversity to the STEM pipeline; has linked researchers and students from across the US and around the world; and has been the basis for extensive education and outreach programs to the K-12 community and the public at large. The JdF major program has explored these and related questions:

* What are the rates, directions, and distributions of fluid and coupled energy, solute, and microbial processes within the volcanic crustal aquifer?
* How do microbial populations in the volcanic crust differ from location to location, and from those found in the overlying ocean and sediments?
* How do microbial processes contribute to rock and fluid alteration?

These questions continue to be important in C-DEBI Phase 2, although the STC is now focused more strongly around the three research themes, all of which are being addressed at JdF. In the following summary, we describe how JdF research has helped to elucidate conditions and processes that are important for an end-member of ridge-flank hydrothermal systems, one in which flows are relatively slow, leading to long residence times and extensive water-rock-microbial interaction. By comparing these results to those from the NP and DO regions, C-DEBI is helping to map out the parameter space in which microbial habitats in the deep subsurface develop, evolve, and are sustained. These other sites are more characteristic of younger and less isolated volcanic crust, whereas the JdF region is more typical (on a global basis) of conditions found in moderate to old ocean crust. The JdF is also the location for the first cross-hole, tracer-injection experiment in the ocean crust, which (as discussed below) provides important information on the nature of flow channeling and isolation within heterogeneous volcanic rocks. Lastly, JdF has produced some of the largest high-quality subseafloor fluid samples, which have proven critical for characterization of the crustal biome in this setting.

**Summary of Significant Accomplishments During Review Period**

At-sea work as part of C-DEBI was completed in the JdF region in 2014, and subsequent research has focused on samples and data derived from earlier studies. There have been proposals submitted to return to this region in future years, but the current focus is on leveraging results from materials and information gathered earlier. The JdF major program has continued to produce important results in 2016, with 5 new studies published in peer-reviewed journals, 1 PhD dissertation, and 21 oral and poster presentations at national and international meetings. Numerous researchers, graduate students and postdocs remain involved in JdF studies, and additional work is underway. Selected research results are highlighted in the following paragraphs, with an emphasis on peer-reviewed publications.

The first three-dimensional simulations of coupled circulation within a ridge-flank hydrothermal system was published in *Nature Communications* last year(Winslow and Fisher, 2015), and a more comprehensive study was published this year in the *Journal of Geophysical Research* (Winslow et al., 2016). Both contribute predominantly to Theme 1. The focus of these studies is properties and processes controlling the dynamics and sustainability of an outcrop-to-outcrop hydrothermal siphon capable of transporting heat and solutes (and perhaps microbes?) across distances of tens of kilometers within the upper ocean crust. In the JdF region, field studies have helped to identify one large seamount that is a site of hydrothermal recharge, and several others that are sites of hydrothermal discharge. Winslow et al. (2016) assess crustal permeability and aquifer thickness, outcrop permeability, the potential influence of multiple discharging outcrops, and differences between two-dimensional (profile) and three-dimensional representations of the natural system. Field observations that help to constrain new simulations include a modest range of flow rates between recharging and discharging outcrops (~5-20 kg/s), secondary convection adjacent to the recharging outcrop, crustal permeability determinations made in boreholes, and the lack of a regional seafloor heat flux anomaly as a consequence of advective heat loss from the crust. Three-dimensional simulations are most consistent with field observations when models use a crustal permeability of 3 x 10-13 to 2 x 10-12 m2, a relatively narrow range that is below estimates based on earlier modeling studies, but consistent with borehole measurements. In addition, models are most consistent with observations when the crustal aquifer is ≤300 m thick. Modeling shows fluid flow rates and crustal cooling efficiencies that are an order of magnitude greater in three-dimensional simulations than in two-dimensional simulations using equivalent properties. Simulations that include discharge from an additional outcrop can also replicate field observations but tend to increase the overall rate of recharge and reduce the flow rate at the primary discharge site. The new models also help to explain the complex geometry of secondary convection near both recharge and discharge sites.

A study based on the first cross-hole tracer experiment in the volcanic ocean crust was published during the 2016 review period (Neira et al., 2016). This study, contributing in particular to Theme 1, tracked the injection and transport of a dissolved gas tracer (sulfur hexafluoride, SF6), using multiple CORKs instrumented with autonomous fluid samplers. All of the fluids used for this study were collected using wellhead samplers, with tubes and fittings that connected the samplers to crustal intervals hundreds of meters below the seafloor (additional work is underway with samples recovered from subseafloor samplers). During the first three years after tracer injection, SF6 was transported both north and south through the basaltic aquifer, with an observed tracer transport rate of ~2-3 m/day. This transport velocity is orders of magnitude faster than inferred from either thermal and chemical observations or calculated as the volume/area rate of fluid transport based on three-dimensional numerical simulations (described above). Taken together, these results suggest that the effective porosity of the upper volcanic crust, the volumetric fraction of rock through which most lateral fluid flow occurs, is <<1%, with most of the fluid flowing rapidly along a few well-connected channels, and the rest of the crust being dominated by diffusive and reactive processes. This finding is consistent with the heterogeneous (layered, faulted, and/or fractured) nature of volcanic upper ocean crust, and will require careful representation of physical and geochemical characteristics in future reactive transport models (e.g., relatively low specific surface area for reaction, limited interaction between microbes and flowing fluids). Additional work is underway to analyze tracer concentrations in downhole samples recovered in Summer 2014 from four CORK systems. These samples provide critical information on tracer transport during the four years following tracer injection, including a time period that is not represented in the seafloor samples analyzed to date, prior to wellhead instrumentation of new CORKs installed on IODP Expedition 327.

Jungbluth et al. (2016) analyzed 1.7 million small subunit ribosomal RNA genes amplified and sequenced from marine sediment, bottom seawater and basalt-hosted deep subseafloor fluids that span multiple years and locations on the JdF. These data, which align closely with Theme 2, delineate a subseafloor microbiome comprised of distinct bacteria and archaea. Hot, anoxic crustal fluids tapped by newly installed seafloor sampling observatories at two boreholes (U1362A and U1362B) contained abundant bacterial lineages of phylogenetically unique Nitrospirae, Aminicenantes, Calescamantes and Chloroflexi. Although less abundant, the domain Archaea was dominated by unique, uncultivated lineages of marine benthic group E, the Terrestrial Hot Spring Crenarchaeotic Group, the Bathyarchaeota and relatives of cultivated, sulfate-reducing Archaeoglobi. Consistent with recent geochemical measurements and bioenergetic predictions, the potential importance of methane cycling and sulfate reduction were imprinted within the basalt-hosted deep subseafloor crustal fluid microbial community. This unique window of access to the deep ocean subsurface basement reveals a microbial landscape that exhibits previously undetected spatial heterogeneity.

Baquiran et al.(2016) explored microbe-mineral interactions within the JdF volcanic oceanic crust, to evaluate microbial colonization on minerals that were incubated in borehole fluids for 1 year at a CORK wellhead (U1301A), and compared these results to a longer-term, downhole experiment in another hole (U1301B) just 36 m away. In comparison to earlier work, this approach, responding to Theme 2, allowed an assessment of temperature, fluid chemistry, and mineralogy on microbial colonization patterns, and allowed verification of the approach of deploying experimental systems at wellheads (which is easier and more versatile in terms of system design, space, etc.). The deployment at U1301B did not result in biofilm growth, based on microscopy and DNA extraction, confirming that this system was not influenced by intrusion of bottom seawater. In contrast, the U1301A installation supported biofilms dominated by Epsilonproteobacteria and Gammaproteobacteria (~44% and ~29%, respectively, of 370 16S rRNA gene clone sequences). Sequence analysis shows overlap in microbial communities that colonized different minerals deployed on the Hole U1301A wellhead, showing that mineralogy did not separate biofilm structure within the one-year experiment. Differences in the U1301A wellhead biofilm community composition relative to that found in previous (downhole) studies, using similar mineral substrate, suggests that the temperature of bottom water and/or the diffusion of dissolved oxygen through the plastic housing influenced the mineral colonization experiments. This work shows the capacity of low-abundance crustal fluid taxa to rapidly establish communities on various mineral substrates under dynamic environmental conditions, and emphasizes the value (despite greater technical difficulty and higher operational cost) of downhole studies.

Robador et al.(2016) completed a nanocalorimetric study as part of Theme 2 to quantify the energy needs of microbial populations recovered from the volcanic ocean crust. The system used has extremely high sensitivity (down to 1.2 nW ml-1), and measured the enthalpy of microbially catalyzed reactions as a function of temperature in samples from two distinct crustal fluid aquifers. Microorganisms in unamended, warm (63°C) and geochemically altered anoxic fluids taken from 292 meters sub-basement (msb) near the JdF produced 267.3 mJ of heat during a 97 h step-wise isothermal scan from 35.5 to 85.0°C. Most of this heat signal likely resulted from germination of thermophilic endospores (6.66 × 104 cells ml-1FLUID) and their subsequent metabolic activity at temperatures greater than 50°C. The average cellular energy consumption (5.68 pW cell-1) reveals the high metabolic potential of a dormant community transported by fluids circulating through the ocean crust. By contrast, samples taken from 293 msb from cooler (3.8°C), relatively unaltered oxic fluids at NP, produced 12.8 mJ of heat in a 14 h experiment, as temperature ramped from 34.8 to 43.0°C. Corresponding cell-specific energy turnover rates (0.18 pW cell-1) were converted to oxygen uptake rates of 24.5 nmol O2 ml-1FLUID d-1, validating previous model predictions of microbial activity in this environment. Given that the investigated fluids are characteristic of expansive areas of the upper oceanic crust, the measured metabolic heat rates can be used to constrain boundaries of habitability and microbial activity in the oceanic crust.

Bach(2016) also explored the energetics of crustal microbial organisms, comparing equilibrium thermodynamic computations to results for kinetic reaction paths. Environmental constraints for this study, closely aligned with Theme 1, were provided by data from several field sites, including the JdF. In these calculations, it was assumed that dissolution of olivine and basalt glass controlled the rates of hydrogen forming reactions in ultramafic and basaltic rocks, respectively. The results suggest that all ocean crust basement rocks release enough hydrogen to support hydrogenotrophic life at low water-to-rock ratios. Olivine dissolution rate control imposes a stronger effect on hydrogen production than phase equilibrium controls, indicating that magnetite formation is not a requirement for production of large amounts of hydrogen in ultramafic rocks. The formation of nontronite and celadonite are primarily responsible for the formation of the moderate amounts of hydrogen expected in basaltic ridge flanks. Under conditions of large seawater fluxes required to account for the great global convective heat flow in ridge flanks, however, hydrogen production in basaltic ridge flanks is insufficient for supporting hydrogenotrophic life. On this basis, it was proposed that the role of Fe oxidation in basaltic ridge flanks is greater than previously thought. A standing stock of 2.4 x 1028 cells could be supported by Fe oxidation in basaltic ridge flanks, equivalent of about 10% of the sedimentary deep biosphere. The size of a hydrogenotrophic biomass within the ocean crust is more difficult to estimate because the rates and processes of hydrogen release are insufficiently constrained. In any case, hydrogenotrophy in the ocean crust could be of key importance mainly in olivine-rich basement rocks and in sedimented ridge flanks with low time-integrated seawater fluxes.

The studies described above, other results presented at meetings, and additional work in progress, could not have been completed without large volumes of pristine fluids from deep below the seafloor. C-DEBI made collection of these materials possible. Developing and applying linked tools that permit access to samples of this kind (clean CORK observatory sealing and sampling systems, long-term pumps with inline filters, autonomous instrumentation control, etc.) has profoundly influenced understanding of the deep, subseafloor biosphere. These studies also show how the primary research themes are being addressed at individual field sites, and by comparison of results at multiple sites. Thermal, tracer, and modeling studies help to understand environmental context, contributing to Theme 1 (*Fluxes, Connectivity, and Energy)*. In addition environmental, laboratory, and theoretical studies are helping to resolve questions related to Theme 2 (*Activities, Communities, and Ecosystems*) and Theme 3 (*Metabolism, Survival, and Adaptation*).Additional work is underway, including synthesis and modeling studies, helping to leverage results from the JdF field region.

► See more at the [Juan de Fuca Ridge Field Site webpage](http://www.darkenergybiosphere.org/research-activities/field-sites/#1470433554305-eeec91f1-4830)

► See References Cited in [[Appendix A](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-A-References-Cited.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2015/12/Appendix-A-References-Cited.pdf)

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**b. Field Site: South Pacific Gyre**

**Led by:** Steven D’Hondt, University of Rhode Island

**Background**

The focus at this study site is on life beneath the seafloor in the most oligotrophic region of the world ocean - the South Pacific Gyre (SPG). IODP Expedition 329*,* led by Co-chief Scientists Steven D’Hondt and Fumio Inagaki, cored and logged deep-sea sediment and basaltic basement at seven SPG sites in 2010. Our present activity in this program focuses on post-expedition studies of samples and data from that expedition. The primary purposes of this project are to:

* Document the nature of microbial communities and test the energetic limit to life in the most food-poor deep-sea sediment,
* Test the influence of basement age and sediment thickness on basement habitability, microbial communities, and the hydrologic evolution of crustal basalt.

This project addresses fundamental questions about subseafloor life, including the following: Is there a lower limit to life in oligotrophic subseafloor sediment? Are the communities in mid-gyre subseafloor sediments uniquely structured (how do these communities compare to those previously studied nearer to the continents)? Is the primary electron donor organic matter from the surface world or hydrogen from *in situ* radioactive splitting of water? Do microbial activities and composition vary with properties of the surface world, such as sea surface chlorophyll concentrations or organic flux to the seafloor? Is microbial activity sustainable in subseafloor basalt by mineral oxidation (*e.g.,* oxidation of iron in the basaltic minerals) or other processes for tens of millions of years after basalt formation? Answering these questions will help to achieve all of the broader C-DEBI themes/objectives. The fundamental goals, activities and outcomes from this reporting period have not different substantially from those originally proposed.

**Summary of Significant Accomplishments During Review Period**

In 2016, we built on our SPG discoveries by expanding our studies of radiolytic H2 production to include North Atlantic and North Pacific abyssal clay provinces. Our results indicate that radiolytic H2 is the primary electron donor available to microbes in sediment older than a few million years in the abyssal clay of all three ocean basins (Sauvage et al., in prep).

The SPG program, which aligns most closely with Theme 2, had several additional scientific accomplishments in this project year. These include (i) experimental demonstration that H2 production per unit of radiation is amplified by up to a factor of 33 in abyssal clay (Sauvage et al., 2016), (ii) demonstration that bacterial diversity (richness of 97%-similar 16S tags) in anoxic subseafloor sediment declines exponentially with sediment age, in parallel with total community respiration (Walsh et al., 2016), (iii) demonstration that some marine microfossil DNA survives for more than one million years (Kirkpatrick et al., 2016), (iv) demonstration that radiolytic H2 may be the primary electron donor in fractures of SPG basalt older than 10 Ma (Dzaugis et al., 2016), and (v) publication of the contamination-tracing results for IODP Expedition 329 (the SPG drilling expedition; Sauvage et al., in press).

The first result indicates that (i) radiolytic H2 is an especially important electron donor for microbes in organic-poor abyssal sediment, and (ii) zeolite should be used with caution (or not at all) in treatment of major radioactive spills (such as the Fukushima disaster). The second result indicates that taxonomic selection in subseafloor sedimentary communities is closely linked to the flux of energy to the community. The third result indicates that a fraction of the DNA in this environment survives for an extraordinarily long time. The fourth result demonstrates that radiolytic H2 may be important for microbial survival in ancient (>10 Ma) subseafloor basalt. The fifth result provides necessary context for all studies of the SPG microbial ecosystem that use Expedition 329 samples or data.

► See more at the [South Pacific Gyre Field Site webpage](http://www.darkenergybiosphere.org/research-activities/field-sites/#1470433554407-0c90ef5c-cd18)

► See References Cited in [[Appendix A](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-A-References-Cited.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2015/12/Appendix-A-References-Cited.pdf)

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**c. Field Site: North Pond**

**Led by:** Geoff Wheat, University of Alaska at Fairbanks

**Background**

The North Pond (NP) project investigates the origin, nature, and activity of microbial communities within basaltic basement below an isolated sediment "pond". About one-third of the seafloor is underlain by oceanic crust that was formed at slow spreading ridges, resulting in seafloor morphology with many ridges and valleys (“mountain ranges”) that are roughly parallel to the spreading center. With age the valleys fill with sediment, but exposed basalt is commonplace, allowing seawater to ventilate and cool the crust. NP is located on the western flank of the Mid-Atlantic Ridge at 22°45'N and 46°05'W and overlies 8 Ma-old crust. Previous exploratory studies there provided a geologic and hydrologic context for microbial studies. Most recently, subseafloor observatories (CORKs) were installed at NP during IODP Expedition 336 (Sept.-Nov. 2011) to collect and monitor active, low temperature, oxygenated fluids that advect vigorously through basaltic basement and to take the first step in conducting manipulative experiments within this natural setting. The three guiding questions for research at NP are:

* What is the nature of microbial communities harbored in young ridge flanks and what is their role in ocean crust alteration?
* Are these communities unique, particularly in comparison with seafloor and sedimentary communities?
* Where do deep-seated microbial communities come from (sediment, rock, seawater, other)?

During IODP Expedition 336, material was recovered (sediment and crustal rocks) and three borehole observatories (CORKs) were installed. Five months later (April 2012) an ROV (*Jason II*) expedition deployed another observatory (CORK-Lite), collected fluids from the CORKs, deployed experiments, and recovered pressure data. A second ROV (*Jason II*) expedition in April 2014 collected more CORK fluids, recovered seafloor experiments, and deployed some additional ones. In 2017 a UNOLS ship will return to NP to sample borehole fluids and recover downhole samplers and sensors.

**Summary of Significant Accomplishments During Review Period**

*Scientific Accomplishments*

Research in 2016 continued to focus on analyses of samples from the 2011 drilling expedition

and the 2012 and 2014 ROV programs. Accomplishments span a range of disciplines, using data from sediment, basement rock, and fluid samples, and technical advances that have been developed by C-DEBI investigators. Key results from a selection of published articles and manuscripts in review are summarized here, with major contributions to Theme 2 and secondary contributions to Theme 3.

* Meyer et al. (2016) (involving two C-DEBI postdoctoral fellows) published, “A distinct and active bacterial community in cold oxygenated fluids circulating beneath the western flank of the Mid-Atlantic Ridge”.Using data and samples collected from subseafloor observatories they report fluid composition and cell abundances that are similar to bottom seawater. However, the microbial community is distinct from that in bottom seawater, revealing an active, distinct, and diverse bacterial community engaged in both heterotrophy and autotrophy in the oxygenated crustal aquifer.
* Russell et al (2016) (including a C-DEBI graduate student fellow) published, “Deep subsurface life from North Pond: enrichment, isolation, characterization and genomes of heterotrophic bacteria.” They used sediment and underlying basaltic samples from IODP Expedition 336 as an inoculum for enrichment and growth experiments, focusing on nitrogen pathways. This work reports on three isolated aerobic heterotrophs from North Pond sediments, which will form the foundation for further studies into geochemical factors impacting life in the deep subsurface.
* Robador et al. (2016) (involving two C-DEBI postdoctoral fellows) published “Nanocalorimetric characterization of microbial activity in deep subsurface oceanic crustal fluids.” They used a nanocalorimeter to measure the enthalpy of microbially catalyzed reactions as a function of temperature in samples from JdF and NP. The NP samples, when converted to oxygen uptake, consumed dissolved oxygen at a rates of 24.5 nmol O2 ml-1FLUID d-1, validating previous model predictions of microbial activity in this environment.
* Zhang et al. (2016) published, “Nitrogen stimulates the growth of subsurface basalt-associated microorganisms at the western flank of the Mid-Atlantic Ridge.” They report enrichment and growth experiments utilizing different carbon (bicarbonate, acetate, methane) and nitrogen (nitrate and ammonium) sources utilizing basaltic materials recovered from IODP Expedition 336. Cell growth was stimulated with nitrogen substrates, and microbial iron oxidation is supported as an important process for microbial communities in subsurface basalts.
* Jørgensen and Zhao (2016) published, “Microbial inventory of deeply buried oceanic crust from a young ridge flank.” They examined 33 basaltic samples collected on IODP Expedition 336. Cell number in these samples are relatively stable at ~104 per gram of rock. The microbial communities in these basalts are distinct from the overlying sediment, yet many of the respective microbial inhabitants are shared between the sediment and basaltic biomes, but with markedly different relative distributions.
* Harigane et al. (2016) published, “Melt-rock interactions and fabric development of peridotites from North Pond in the Kane area, mid-Atlantic ridge: implications of microstructural and petrological analyses of peridotite samples from IODP Hole U1382A.” They analyzedperidotite and gabbro samples from IODP Expedition 336. Petrographic studies are consistent with the reinvigoration of mantle melt that occurred in an extinct spreading segment after spreading at the axis and subsequent movement off axis. This highlights the potential for sill formation off axis and the potential to affect hydrologic properties of the crust.

*Technical Accomplishments*

The NP program continues to push technology in areas that serve the development of borehole

installations for experimentation. We are currently developing an in situ flow meter that worked for a short period in the JdF CORKs for use in the return to NP in 2017 and improving the “harpoon” in hopes for recovering the borehole samplers in Hole 395A, which lost the wellhead upon deployment but retains the boreholes packages that could be recovered with a harpoon system. We also are improving the manipulator-triggered syringe-water sampling systems that were developed for the DO site. Lastly CORK-Lites, which were developed for NP boreholes, are being exported for use in four boreholes that are currently being deployed on IODP Expedition 366 to the Mariana Forearc Serpentinite Mud Volcanoes (December 2016 - February 2017).

► See more at the [North Pond Field Site webpage](http://www.darkenergybiosphere.org/research-activities/field-sites/#1470441369069-3559f6eb-f4fd)

► See References Cited in [[Appendix A](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-A-References-Cited.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2015/12/Appendix-A-References-Cited.pdf)

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**d. Field Site: Dorado Outcrop**

**Led by:** Geoff Wheat, University of Alaska at Fairbanks

**Background**

This study builds on decades of research that observed differences between measured and theoretical heat flow values in the ocean crust. Lister (1972) postulated that these differences resulted from the cooling of the crust by the circulation of seawater. The magnitude of this circulation is large, redistributing 10 TW (one fourth) of the Earth’s heat loss. Then, in 1977, the first seafloor hydrothermal system was discovered; however, such systems, driven by the intrusion of magma, account for only 20% of the total convective heat loss from oceanic crust. The remaining heat loss is transported on the ridge flanks at much cooler temperatures, yet with a net fluid flux that is commensurate with that of rivers. Given this magnitude of fluid flow, it has been postulated that even a minimal (1-5%) loss or gain from seawater-crustal exchange could impact global geochemical budgets in the ocean; however, until recently such a representative system had yet to be sampled.

Off the west coast of Costa Rica on 23 Ma-old crust lies the Dorado Outcrop (DO), a small (0.5 km wide by 2 km long and 150 m high) basaltic outcrop that trends southeast-northwest and is characteristic of typical ridge flank hydrothermal systems. In 2013, we surveyed DO using the autonomous underwater vehicle Sentry and the remotely operated vehicle Jason II (AT26-09). Surveys generated a bathymetric map from which visual and thermal surveys located sites where crustal fluids discharge from the crust. In 2014, we returned to DO with the submersible Alvin with which we recovered samplers and sensors, collected high-quality fluids, and measured dissolved oxygen in situ.

**Summary of Significant Accomplishments During Review Period**

*Scientific Accomplishments*

In 2016, we built upon the operational success from 2013 and 2014 through the analysis of kilometers of high resolution bathymetric, discrete and continuous spring and background fluid samples, sediment and associated pore fluids, rocks, heat flow measurements, and in situ temperature and dissolved oxygen. These analyses are being incorporated into a number of manuscripts, one of which is published.

One submitted manuscript highlights some aspects of the spring fluid composition and aligns with Theme 1. Analysis of these fluids reveals that discharge from cool ridge-flank systems on a global basis could result in fluxes of Rb, Mo, V, U, Mg, phosphate, and Li that are ≥10% of the riverine flux. In addition, spring fluids have ~50% less dissolved oxygen than bottom seawater. This oxygen loss occurs primarily within the basaltic crust, demonstrating that (1) permeable pathways within the upper crust can remain oxic for millions of years, and (2) that reactions within the crust consume dissolved oxygen. The published paper (Lee et al., December 2015) examined microbe-mineral interactions on rocks that were recovered from areas bathed in these spring fluids. Their results, targeting Theme 2, reveal a much greater richness and diversity on these rocks than the surrounding seawater. The most abundant bacterial reads were closely linked to obligate chemolithoautotrophs. Results from this study coupled with similar studies elsewhere suggest cosmopolitan phylogenetic groups and that substrate age correlates with community structure.

A third manuscript goes beyond these local studies to elucidate regional subsurface hydrologic flow. This manuscript, contributing to Theme 1, builds upon three-dimensional models of subsurface hydrothermal circulation that were initially developed to simulate flow through the crust on the eastern flank of JdF. The current manuscript examines the effective flow, pathways, and potential residence time for individual pathways through a seamount network that is representative of the DO area and fast-spreading crust in general. Combined with the other two manuscripts, these works demonstrate the potential for cool ridge-flank hydrothermal systems to influence crustal alteration, the subseafloor biosphere, and geochemical budgets of the global ocean.

The samples and data that were collected are and will be incorporated into additional manuscripts, including:

* Fisher et al. (heat flow and water column thermal anomalies) – Systematic variations in heat flow measurements coupled with estimates of sediment thickness from chirp sonar data constrain the total flux of heat and fluid that vent from the outcrop. Much of this flow was visualized during the two field campaigns.
* Wheat et al. (geology and flow) – Fluid flow from DO is focused through pillow basalt structure. This flow is tidally forced, resulting in variations in fluid flow rates with time.
* Hartwell et al. (octopus) – Deep-sea octopi thrive in areas of warm fluid seepage and lay their eggs within the spring flow, contrary to the theory that such flow would harm both mother and eggs.
* Bach et al. (secondary mineral alteration) – Twenty million years of exposure to bottom seawater coupled with millions of years of crustal fluid flow have led to alteration products within basalts recovered from the seafloor. Alteration products reflect the oxygenated and minimally altered seawater that flows through the oceanic crust at this location.
* McManus et al. (organic chemistry of fluids) – Formation fluids that vent at the seafloor have decreased dissolved inorganic carbon concentrations relative to bottom water. Dissolved organic carbon concentrations also are lower, pointing towards consumption within the oceanic crust.

*Technical Accomplishments*

A combination of the fluid sampling techniques that were developed for DO and the technical accomplishments that were developed during the tracer experiment on the JdF eastern flank provide the foundation for a tracer study in the DO region. Here fluid flow through the crust is fast, likely an order of magnitude or more faster than on the eastern flank of JdF. This is an ideal site to develop such a study of crustal hydrologic properties, seawater-rock exchange, and microbial processes in oceanic crust formed from a fast spreading center. As a result of technical and scientific accomplishments we will develop a drilling proposal for this site to be submitted in April 2017.

► See more at the [Dorado Outcrop Field Site webpage](http://www.darkenergybiosphere.org/research-activities/field-sites/#1470441918956-18567177-03b5)

► See References Cited in [[Appendix A](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-A-References-Cited.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2015/12/Appendix-A-References-Cited.pdf)

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**e. Field Site: Atlantis Massif**

**Led by:** Beth Orcutt, Bigelow Laboratory for Ocean Sciences

**Background**

The Atlantis Massif (AM), a new focus area of C-DEBI, is an ocean core complex of mantle-type oceanic crust uplifted to the seafloor on the western flank of the Mid-Atlantic Ridge (a few degrees latitude north of NP). Unlike the basaltic crustal systems at JdF and NP, this ultramafic, mantle-type rock undergoes alteration called serpentinization when exposed to seawater, where the original ultramafic igneous rocks are hydrated and metamorphosed into serpentinites. Chemical byproducts of the serpentinization reaction include abiotic generation of hydrogen and small carbon compounds, an increase in pH, and a lowering of alkalinity. AM is the type site for studying such reactions on Earth, as it hosts the spectacular Lost City hydrothermal vent field discovered less than 20 years ago, characterized by 60-m-high towers of carbonate chimneys and venting fluids. This style of hydrothermal venting is hypothesized to also occur on other ocean worlds where ultramafic rocks are present.

C-DEBI scientists, working with international colleagues, were involved in a proposal to the IODP to drill into AM to understand the process of serpentinization, and how a deep biosphere in this environment would compare to that hosted in basaltic systems or to serpentinization environments on land. Unlike traditional ocean drilling programs using a drill ship, this expedition used seabed drills to enable the collection of intact sequences of highly heterogeneous and altered rocks from the upper tens of meters of the Massif – such high recovery would not be possible with conventional drilling. Moreover, the proposed program developed new technologies for the seabed drills – such as water samplers, chemical sensor packages, wireline logging tools, tracer delivery systems, and borehole plug systems – to enable contextual analysis of the rocks recovered. The primary, interdisciplinary goals of the proposed work are to: (1) examine the role of serpentinization in driving hydrothermal systems, sustaining microbial communities, and sequestering carbon; (2) characterize the tectonomagmatic processes that lead to lithospheric heterogeneities and detachment faulting; and (3) assess how abiotic and biotic processes change with variations in rock type and progressive exposure on the seafloor. Specific deep biosphere hypotheses to be tested are:

* An extensive subsurface hydrogen-based biosphere persists in actively serpentinizing lithosphere. These communities evolve and adapt to variations in diffuse fluid flow, fluid chemistry, heat flow distribution, and age of exposure on the seafloor.
* Serpentinizing environments sustain higher biomass than gabbroic-dominated domains.
* The transition from sulfide- to carbonate-dominated environments can be detected by changes in the rare biosphere of the associated microbial communities.
* Zones of intense carbonate veining underlie sites of diffuse venting and represent seawater recharge and net sequestration of CO2 from the hydrosphere into the lithosphere. These zones are biological hot spots where microbial communities are supported by the high fluxes of hydrogen mixing with carbon dioxide.

These hypotheses are directly connected to the aims of all three research themes (*Fluxes, Connectivity, and Energy; Activities, Communities, and Ecosystems; Metabolism, Survival, and Adaptation*).

**Summary of Significant Accomplishments During Review Period**

C-DEBI Senior Scientist Beth Orcutt served as one of the Co-Chief Scientists of Expedition 357, and several C-DEBI supported scientists (Billy Brazelton, Susan Lang, Matthew Schrenk, Katrina Twing) were members of the offshore and onshore international science parties. The offshore phase took place from October 26, 2015 – December 11, 2015, followed by the onshore phase for cutting and describing the core in Bremen, Germany, from January 20 – February 4, 2016. Expedition 357 successfully used seabed drills for the first time in the ocean drilling program, recovering 57.09 meters of core from 17 holes drilled at 9 sites across the massif, with core recoveries as high as 75% in some cases (Früh-Green et al., 2016). This high level of recovery of shallow mantle sequences is unprecedented in the history of ocean drilling. In addition, new technologies were used successfully on the expedition, namely (1) extensively using an *in situ* sensor package and water sampling system on the seabed drills for evaluating real-time dissolved oxygen and methane, pH, oxidation-reduction potential (ORP), temperature, and conductivity during drilling; (2) deploying a borehole plug system for sealing seabed drill boreholes at two sites to allow access for future sampling; and (3) proving that chemical tracers for contamination testing can be delivered into drilling fluids when using seabed drills. A major achievement of Expedition 357 was obtaining samples for microbiological analysis, which will provide a better understanding of how microbial communities evolve as ultramafic rocks are emplaced on the seafloor.

Initial data from the Expedition shows that wide-scale, active serpentinization is on-going at AM, as indicated by recordings of the sensor packages and by elevated concentrations in H2 and CH4 in bottom water sampled before and after drilling. Monitoring of the borehole fluids during drilling operations recorded numerous excursions in methane, temperature and ORP that often correlated with each other, implying that horizons of hydrogen-rich fluids must exist in the basement rocks, and that volatiles are being continuously expelled during active serpentinization at AM. Active volatile expulsion was also visible as bubbles emitting from the two most western sites. These results indicate that the subsurface of the serpentinite basement of AM provides a potentially important niche for anaerobic hydrogen- and methane-cycling microorganisms.

During the offshore phase, C-DEBI scientists set up numerous experiments on the ship to examine various microbial activities and processes, such as methane and sulfur cycling. Following the onshore phase in Bremen, C-DEBI scientists in the science party traveled to Kochi, Japan, in late February 2016 to process the 48 frozen samples collected for microbiological and geochemical analysis using the state-of-the-art clean room facilities at the Kochi Core Center for processing frozen core samples. This collaborative effort was supported through a partnership with the Japanese IODP program and the Sloan Foundation-funded Deep Carbon Observatory Deep Life program. The 48 samples were pooled and subsampled for various coupled analyses by the various deep biosphere scientists in the science party. Analyses are underway in all of these laboratories, and Orcutt organized monthly conference calls to share initial results during the Expedition moratorium period (ending February 2017).

► See more at the [Atlantis Massif Field Site webpage](http://www.darkenergybiosphere.org/research-activities/field-sites/#1470442621363-98e201bc-ec6f)

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**f. Other Field Projects**

In addition to the major field programs reviewed above, C-DEBI is involved in other expedition-based research as well. Here, we briefly highlight four of these projects, identifying some key C-DEBI personnel involved:

***Baltic Sea Basin sediment***. The Center for Geomicrobiology at Aarhus University (Denmark) led a June 2016 cruise to revisit a site previously sampled by IODP Expedition 347 in 2013. This site’s high organic content and high sedimentation rates lead to anoxia at or immediately below the seafloor. Shallow (less than 10 meters below seafloor) sediment cores were taken to understand biogeochemical cycling and microbial communities in this basin. Findings from this ‘shallow’ sediment are being compared to deeper sediment collected by Expedition 347 at the same site. Metatranscriptomics and nucleic acid quantification are being performed on the sediment. Ongoing work on Expedition 347 sediment shows *Proteobacteria*, *Firmicutes*, and *Chloroflexi* dominate the active microbial communities. Genes indicative of fermentation and complex carbohydrate breakdown are the primary catabolic genes expressed. We will compare results from the sediment collected in 2016 to these deeper transcriptional profiles to determine how catabolic gene expression changes with depth. (Graduate Student Laura Zinke, Dr. Brandi K Reese, and Dr. Jan Amend)

***T-Limit of the Deep Biosphere off Muroto***. IODP Expedition 370 (13 September - 11 November, 2016) cored deep beneath the seafloor with the scientific objectives of (i) documenting the thermal limit to subseafloor sedimentary life, and (ii) determining the nature of subseafloor life close to its thermal limit. C-DEBI participants are focusing on (i) geochemical evidence of thermal limits to biological activity, (ii) bioenergetics and (iii) microbe-mineral interactions of this subseafloor ecosystem. (Graduate Student Justine Sauvage, Graduate Student Kyle Metcalfe, Dr. Steven D’Hondt and Dr. Victoria Orphan)

***Mariana serpentinite mud volcanism: geochemical, tectonic, and biological processes***. IODP Expedition 366 departed on December 8, 2016 and will return February 8, 2017. This expedition has two primary science objectives. The first is to core a series of sites at the summit and flanks of three large (up to 50 km diameter and 2 km high) serpentinite mud volcanoes in the Mariana forearc (within 100 km west of the Mariana Trench). This objective addresses the broad scientific aim of examining processes of mass transport within the subduction zone of a nonaccretionary convergent margin. The second objective is to establish long-term seafloor observatories by emplacing cased boreholes at summit (conduit) holes in three mud volcanoes and removing the CORK body from an old ODP hole (1200C). These activities will set the foundation for future deployments of sensors and samplers, e.g., by deployment of CORK-Lite structures within the boreholes, thus providing a framework for conducting temporal observations that will allow one to “take the pulse of subduction” in an active nonaccretionary convergent plate margin and establish a platform for in situ experimentation. (Dr. Geoff Wheat)

***Searching for Life in the Mariana Back-Arc.*** This cruise was from 29 November to 20 December, 2016 aboard the R/V Falkor, with ship and ROV time provided by the Schmidt Ocean Institute and scientific support from NOAA Office of Ocean Exploration and Research. Home of the deepest spot on the planet, the Mariana subduction system serves as a valuable natural laboratory for testing ideas about what governs the distribution of microbes beneath the seafloor at hydrothermal vent systems. The deep trench, shallow to mid-depth volcanic arc, and mid-depth to deep spreading back-arc, provide a wide variety of habitats for research. During a previous mapping cruise, also aboard the Falkor in 2015, three new hydrothermal vents were discovered; one of them is among the deepest vents in the world. The team examined the chemistry and geology of the vents, studied their microbiology, and tested explanations of the substantial biological differences between the volcanic arc and back-arc vents. (Dr. Julie Huber)

► See related C-DEBI Contributed Publications in [[Appendix J](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-J-Center-wide-Outputs.xlsx)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-I-Center-wide-Outputs.xlsx)

**g. Key Laboratory Studies**

While deep-sea sediments contain a significant fraction of the global microbial biomass, in situ growth of these microbes is severely limited if not stalled, due to the absence of catabolic substrates or low nutrient mass transport. However, our general understanding of the physiology of microorganisms has been historically derived from fast growing, terrestrial microorganisms. In Theme 3 (*Metabolism, Survival, and Adaptation*), C-DEBI seeks to investigate slow-growing organisms under laboratory conditions and to study their metabolisms in controlled environments that allow for scientific hypotheses testing. Here, we discuss several such ongoing experiments.

***Characterization of Halomonas strains isolated from North Pond***. Using a very low carbon medium, Huber’s group isolated about 40 strains of the bacterium *Halomonas* and then sequenced their 16S rRNA genes. Finkel’s group is now characterizing the suitability of these organisms with respect to their utility as model organisms for laboratory experiments. Of particular interest are the broad metabolic capacities of these organisms, which can survive in media from the most nutrient restrictive to highly rich. Further, the characterization of these strains during the transition from low to high nutrient status indicates that different isolates may be sensing and responding to nutrient stress differentially, providing insight in the different mechanisms used by these highly related microbes.

***Physiology of Dehalococcoides mccartyi, a close relative of deep-sea* *Chloroflexi.***  To study the physiology of very slow growing microorganisms (doubling times of 50 days or longer), the Spormann lab developed parallel anaerobic reactors, where the growth rates of the organisms are tightly constrained via the dilution rate. Questions such as: how long do slow-growing organisms take to replicate their genome? how is their cell-cycle impacted by growth-rate? and what is the limiting factor during DNA replication? have so far remained unresolved. The culture platform in the Spormann lab is well-suited to start answering these questions. Using the parallel reactors, the Spormann lab will study the impact of increasing growth rate on cell cycle, visualizing the distribution of chromosomal content in the cell population via flow cytometry. The Spormann lab is working with a highly enriched *Dehalococcoides mccartyi* culture, which, although isolated from a terrestrial environment, is closely related to deep-sea *Chloroflexi*. Its fastest doubling time is 2-3 days under ideal batch condition, but it grows much more slowly in the environment; in the Spormann lab, it is adapted to a 25 day doubling time, and it can survive extremely low substrate flux. This experimental set-up enables the investigation of the metabolism of slow-growing organisms, with a particular focus on a) how the proteome of a cell responds to starvation, and b) the dynamics of the cell cycle at slow growth rate.

The survival of deep-sea microorganisms on low nutrient availability or complete starvation depends on the resilience and stability of their protein content: if proteins remain stable over long periods of starvation, the cells are equipped to start metabolizing substrates as soon as they return. One goal is to study the impact of extended period of starvation on the *D. mccartyi* cultures and understand the proteome response at the onset of starvation, during starvation and as substrates are made available again. Using shotgun proteomics, the Spormann lab is identifying which proteins are present. For specific catabolic proteins of interest, they will also track the absolute number of proteins per cells using quantitative proteomics. Finally, using the BONCAT technique, the Spormann lab will also monitor which proteins are preferentially synthesized when substrates are returned. Preliminary results show that BONCAT signal in *D. mccartyi* is low but detectable and that protein synthesis resumes within 4h of substrate addition after starvation.

***Growth and long-term survival of Pseudomonas aeruginosa in different pore waters***. The D’Hondt and Finkel groups are collaborating to identify readily metabolizable components in pore waters obtained from a variety of sediment samples. Initial experiments are using sediment pore waters from different depths of the New England Margin. Because of its broad metabolic capabilities, the cosmopolitan heterotroph *Pseudomonas aeruginosa* is being used as the initial bacterial test strain. Preliminary studies have shown that while some pore waters can support substantial microbial growth, more than half contain factors that either inhibit growth or reduce cell viability. Current experiments are targeting the identity of these factors.

**h. Key Modeling Studies**

# When data are sparse and the environment is vast, modeling efforts can facilitate our understanding of the biogeochemical drivers that govern deep life. C-DEBI modeling efforts target both regional and global processes. Applications of regional fluid flow modeling at JdF and DO were described in Sections 2.a and 2.d, respectively. Here, we highlight progress made in 2016 on better understanding the global sediment biosphere; the majority of this work has come out of the Amend lab in collaboration with Doug LaRowe and addresses topics in Theme 1.

# In a *Microbe* Feature Article, Amend and LaRowe (2016) remind a broad microbiology audience that our understanding of the intraterrestrials and their host environments, especially beyond the near-coastal regimes, remains limited and reliant on the analysis of relatively few samples. The communication describes the difficulty in determining whether microbes are active, dormant, or dead, tackling the role played by spores in low-energy sedimentary ecosystems. The article also reviews our knowledge regarding the abundance of archaea versus bacteria, and anaerobes versus aerobes, concluding with a call for more sophisticated energetic and metabolic modeling of life in marine sediments.

# LaRowe et al. (2016) note that marine sediments contribute significantly to global element cycles on multiple time scales, which is due in large part to microbial activity in the shallower layers and abiotic reactions resulting from increasing temperatures and pressures associated with ever deeper burial. Quantifying the rates of these diagenetic changes requires a three dimensional description of the physiochemical properties of marine sediments. In a step towards reaching this goal, this study combined global datasets describing bathymetry, heat conduction, bottom water temperatures and sediment thickness to quantify the 3-D distribution of temperature in marine sediments. This model revealed that significant microbial activity could be inhibited in ~25% of marine sediments, if 80°C is taken as a major thermal barrier for subsurface life. New values for the total volume (3.01 × 108 km3) and average thickness (721 m) of marine sediments were also calculated, providing the only known determination of the volume of marine-sediment pore water (8.46 × 107 km3), equivalent to ~6.3% of the volume of the ocean.

# LaRowe and Amend (2016) quantify the amount of energy required to make biomass as a function of temperature; pressure; redox state; the sources of C, N and S; cell mass; and the time that an organism requires to double or replace its biomass. The amounts of energy associated with synthesizing the biomolecules that make up a cell, which varies over 39 kJ (g cell)-1, are then used to compute energy-based yield coefficients for a vast range of environmental conditions, in particular in marine sediments. The study concludes that, taken together, environmental variables and the range of cell sizes leads to a ~4 orders of magnitude difference between the number of microbial cells that can be made from a Joule of Gibbs energy under the most (5.06×1011 cell J-1) and least (5.21×107 cell J-1) ideal conditions.

# Lastly, in an ongoing study, LaRowe et al. discuss microbial degradation of organic matter in marine sediments on multiple time scales. We posit that it is not known to what depth microorganisms alter organic matter in marine sediments or how microbial rates of organic carbon processing change with depth and time on a global scale. To better understand the connection between the carbon cycle and life’s limits in the deep subsurface, we combined a number of global data sets including sedimentation rates, bathymetry, and particulate organic matter delivery fluxes with a reaction transport model describing organic matter degradation. We then quantify the distribution of organic matter in marine sediments due to microbial degradation. In particular, we estimate the 3-D distribution of particulate organic matter in marine sediments deposited throughout the Quaternary, encompassing the most microbially active layers of marine sediment.

► See related C-DEBI Contributed Publications in [[Appendix J](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-J-Center-wide-Outputs.xlsx)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-I-Center-wide-Outputs.xlsx)

# i. Projects from our Grants and Fellowships Program

As noted above, C-DEBI has an extensive grants and fellowships program, which includes funding opportunities for small research projects, research and travel exchanges, education and outreach, and graduate student and postdoctoral fellowships. The funded projects cut across all three Themes. A list of all 36 funded projects active in 2016 is provided in [[Appendix B](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-B-Active-Grants-and-Fellowships.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2015/12/Appendix-B-Active-Grants-and-Fellowships.pdf). The breakdown of active grants and fellowships is as follows: 4 Phase 1 small research projects with funding up to $50k, 1 Phase 1 special biomolecular grant with funding up to $150k, 6 Phase 1 special synthesis-field programs grants with funding up to $100k, 5 Phase 2 small research projects with funding up to $80k, 6 graduate student fellowships with funding for 1-2 years (4 in Phase 1 and 2 in Phase 2), 10 postdoctoral fellowships with funding for 1-2 years (5 in Phase 1 and 5 in Phase 2), 2 education and outreach grants with funding up to $50k, and 2 research exchange grants that require matching funds. Forty-five different individuals comprised of almost half graduate students and postdocs, representing 26 institutions, received financial support for these projects.

The small research grants and fellowships support a wide variety of field projects, experimental and analytical investigations, and modeling efforts relevant to C-DEBI. These include analyses of samples and data from deep subseafloor sites, laboratory studies of microbial activity, and investigations of analog environments. Here, we call out only the 12 research awards made in 2016. Small research grants (up to $80k) were made to: Ken Nealson (Professor at USC) to investigate the diversity of extracellular electron transfer in deep sea marine sediment; Andrew Steen (Assistant Professor at the University of Tennessee at Knoxville) to assess if hydroxyl radicals liberate bioavailable organic carbon in subsurface sediments; Anne Dekas (Assistant Professor at Stanford) to determine the identities and single-cell activity rates of diazotrophic microorganisms in deep-sea sediments; Susan Lang (Assistant Professor at the University of South Carolina) to investigate the biogeochemistry of fluids from the serpentinite subsurface; and Wiebke Ziebis (Associate Professor at USC) to explore microbial diversity in sediments underlying oligotrophic gyres. Fellowships (up to 2 years support) were awarded to: Tucker Ely (graduate student at Arizona State University) to carry out 3D spatial mapping of the energetic return of 1000 metabolisms within the compositional variation of oceanic crusts near mid-ocean ridges; Emily Estes (graduate student at MIT/WHOI) to assess geochemical controls on organic carbon quantity and quality in the deep subsurface; Blair Paul (postdoc at UCSB) to examine adaptive protein diversification by microorganisms and their viruses in subseafloor sediments using a targeted metagenomic approach; Nagissa Mahmoudi (postdoc at Harvard) to investigate the bioavailability and degradation of sedimentary organic matter; James Bradley (postdoc at USC) to develop a 1D biogeochemical-evolutionary model for deep sediments; Jacqueline Goordial (postdoc at Bigelow) to investigate the diversity and function of active microbial subpopulations in Atlantis Massif oceanic crust; and Gustavo Ramirez (postdoc at URI) to study how to discriminate detrital genes from marine sediments.

► See more at the [Funded Projects webpage](http://www.darkenergybiosphere.org/?s=&type=award)

► See related C-DEBI Contributed Publications in [[Appendix J](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-J-Center-wide-Outputs.xlsx)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-I-Center-wide-Outputs.xlsx)

# j. C-DEBI Workshops

C-DEBI funded and organized two workshops in 2016 to assemble the community around pertinent topics in deep biosphere research. The first one was co-sponsored by the Deep Carbon Observatory Deep Life Community and focused on the origins and movements of subsurface microbes. It was held at USC May 9-10 and was led by Tom Kieft (New Mexico Tech) and Doug LaRowe (USC). The second workshop, held December 10-11 in proximity to the American Geophysical Union Annual Meeting and organized by Ben Tully (USC), was a second annual workshop for hands-on training in bioinformatics.

► See more at the [2016 Workshops webpage](http://www.darkenergybiosphere.org/outputs-resources/meetings-and-workshops/#2016-workshops)

## 3. Performance With Respect to the Strategic Implementation Plan

Our primary research goal is to enable, produce, and communicate transformative, synergistic research through an inclusive and collaborative culture that crosses disciplinary and institutional boundaries and is embedded throughout the Center’s activities. In Phase 1, C-DEBI focused primarily on the exploration and discovery of subseafloor ecosystems, with most studies concentrated at four major sites: Juan de Fuca Ridge flank, South Pacific Gyre, North Pond, and the Dorado Outcrop. In Phase 2 (2015-2020), C-DEBI is developing an integrated understanding of microbial subseafloor life, covering and connecting the molecular, cellular, and ecosystem scales. Maintaining highly multidisciplinary and interdisciplinary approaches, C-DEBI will emphasize microbial ecology while ensuring that essential context is provided through studies and advances in geochemistry, hydrology, oceanography, and related disciplines. The three overarching research themes are (1) fluxes, connectivity, and energy; (2) activities, communities, and ecosystems; and (3) metabolism, survival, and adaptation. C-DEBI research projects target two distinct subseafloor biosphere environments—the igneous ocean crust and overlying sediments—that have historically been studied independently; field investigations of these environments are complemented by coordinated laboratory studies and modeling. C-DEBI is led by five Co-PIs and five senior scientists from eight U.S. universities and research labs, but seeks to build and leverage scientific, educational, and technological partnerships with numerous other U.S. and international institutions (educational, research, outreach, engineering, not-for-profit). In addition, C-DEBI seeks to develop a diverse community of multidisciplinary collaborators, to identify promising topics, and to develop new projects that will help to advance the Center's objectives.

**Target 1:** Transfers of fluid, heat, solutes, carbon, and microbes are quantified within and between subseafloor biomes, and between the subseafloor and the overlying ocean; the nature of energy sources available to microbes in these ecosystems is determined; and the next generation of coupled fluid-energy-biochemical-microbial models is developed.

|  |  |
| --- | --- |
| **Metric** | **Status/Problems** |
| Quantify transfers of fluid, heat, solutes, carbon, and microbes within and between subseafloor biomes, and between the subseafloor and the overlying ocean   1. Continue time-series observations and sampling at selected sites and analyze data and samples from earlier studies to resolve the extent of natural variability (within and between biomes), determine environmental controls on crustal microbial community composition, and assess how this variability impacts flows and connections 2. Develop studies, including some at new field sites, as needed to test and extend understanding of coupled fluid-rock-geochemical-microbial systems | Pending |
| Determine the nature of energy sources available to microbes in subseafloor ecosystems   1. Map the distributions of electron acceptors and electron donors regionally and globally as a function of depth at a range of spatial scales. 2. Quantify metabolic reaction energetics as well as fluxes of electron acceptors and donors by combining internally consistent thermodynamic data, available kinetic parameters, and reactive transport modeling | Pending |
| Develop the next generation of coupled fluid-energy-biochemical-microbial models   1. Combine existing physical and thermal models with rate constants for primary reactions and transport (advection and diffusion) that involve solutes in basement fluids; then attempt to couple these with microbial processes to increase the model complexity 2. Test, calibrate, and apply coupled geochemical-microbiological models to a variety of seafloor and subseafloor environments | Pending |
| Publish 25 (in aggregate) papers in this research theme | Pending |
| Publish 5 (in aggregate) method/instrument papers demonstrating new techniques and tools developed and/or applied in this research theme | Pending |

**Target 2:** The composition of subseafloor microbial communities and the functional potential of these communities are illuminated, based on the diversity of metabolic activities and interactions with the physicochemical aspects of the system.

|  |  |
| --- | --- |
| **Metric** | **Status/Problems** |
| Determine community composition, functional potential, and patterns of natural selection in subseafloor ecosystems   1. Quantify the number, diversity, and relative abundances of microbes at multiple taxonomic levels—from domain to “species-level” operational taxonomic units (OTUs), ecotypes, and oligotypes 2. Determine the functional potential embodied in these communities 3. Integrate data on community composition and microbial activities to identify how sources of energy and microbial interactions drive natural selection in subseafloor ecosystems | Pending |
| Determine metabolic activity of subseafloor microbial communities   1. Document actual rates of in situ activities using gene expression in sediment and rock samples 2. Identify potential activities in laboratory experiments using subseafloor samples incubated with isotope-labeled substrates 3. Closely examine microbe-mineral interactions in conjunction with activity measurements in in situ incubations and laboratory microcosms | Pending |
| Advance understanding of subseafloor microbe-virus interactions   1. Integrate correlation network techniques using subseafloor archaeal, bacterial, microeukaryote, and viral diversity datasets combined with microbial activity measurements 2. Incorporate the isotopic and diversity datasets collected as part of 2.a. and 2.b. to develop a food web model in combination with statistical diversity-based networks | Pending |
| Publish 25 (in aggregate) papers in this research theme | Pending |
| Publish 5 (in aggregate) method/instrument papers demonstrating new techniques and tools developed and/or applied in this research theme | Pending |

**Target 3:** A ‘portfolio’ of selected model subseafloor organisms is established, and their physiological and genetic traits are characterized; in addition, these microorganisms are used to investigate energy and carbon use for growth and maintenance under kinetically limiting conditions and to determine rates of metabolism under specific conditions.

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| --- | --- |
| **Metric** | **Status/Problems** |
| Isolate and characterize novel bacteria and archaea from diverse subseafloor habitats   1. Enrich subseafloor bacteria and archaea from sediment, crustal fluids, and rock samples, using, among others, plugged flow, chemostat, and hanging sponge reactors 2. Fully characterize novel organisms, including their genomes 3. Interrogate their abundance and activity in the original sample to help infer their ecological roles | Pending |
| Examine fundamental physiology of subseafloor microbes under conditions of low growth rates and low energy flux   1. Use long-term chemostat-like culturing systems to study the coupling of catabolism and growth in the Chloroflexi 2. Use down-flow hanging sponge bioreactors to explore the molecular and physiological underpinnings of the hypothesis that archaea may be low-energy specialists and well adapted to the energetic extremes that define many subseafloor environments | Pending |
| Perform adaptive evolution and long-term survival experiments with subseafloor microbes to characterize molecular genetic signatures associated with particular phenotypes   1. Use subseafloor isolates to determine the genotypic, phenotypic, and biochemical and physiological bases for metabolic traits 2. Develop genetic markers for model organisms to be used in competition experiments | Pending |
| Publish 25 (in aggregate) papers in this research theme | Pending |
| Publish 5 (in aggregate) method/instrument papers demonstrating new techniques and tools developed and/or applied in this research theme | Pending |

**Target 4:** Field investigations at the four ‘major sites’ identified in C-DEBI Phase 1 are largely completed (i.e. at Juan de Fuca, South Pacific Gyre, North Pond, and Dorado Outcrop with the latter two potentially continuing beyond C-DEBI Phase 2). Environmental data and samples from these sites are compiled and analyzed along with laboratory experiments and modeling to address questions across the three Phase 2 research themes.

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| --- | --- |
| **Metric** | **Status/Problems** |
| C-DEBI researchers lead and participate in expeditions to these and other sites of interest   1. Collect samples for laboratory analyses and experiments 2. Collect environmental data for use in experiments and ecosystem modeling | Met |
| Convene workshops and conference sessions   1. Develop approaches to integrate results from field, lab, and modeling studies 2. Synthesize results and methods from multiple sites | Met |

**Target 5:** The new C-DEBI senior scientists are integrated in all aspects of the Center, and cross-disciplinary and cross-institutional research training is thriving through our grants programs, thereby expanding the community of deep biosphere researchers, technologists, and educators.

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| --- | --- |
| **Metric** | **Status/Problems** |
| Provide substantial research funds to the 5 Co-Investigator labs as well as to the 5 new senior scientist labs | Met |
| Award $1M in research grants/fellowships annually (for the first 3.5 years) to predominantly graduate students, postdoctoral scholars, and other junior researchers through annual RFPs | Met |

**4. Plans for the Next Reporting Period**

The research plans for the next reporting period remain as stated in our Phase 2 proposal. As outlined in Theme 1, we will constrain the extent, variability, and controls on fluxes and connectivity within subseafloor biomes and between the subseafloor and the overlying ocean; map geochemical energy sources in subseafloor ecosystems at a range of spatial scales; and develop and test the next generation of coupled geochemical-hydrological-microbial models for subseafloor ecosystems. In Theme 2, we will determine community composition, functional potential, and patterns of natural selection in subseafloor ecosystems; determine metabolic activity of subseafloor microbial communities; and advance understanding of subseafloor microbe-virus interactions. In Theme 3, we will isolate and characterize novel bacteria and archaea from diverse subseafloor habitats; examine fundamental physiology of subseafloor microbes under conditions of low growth rates and low energy flux; and perform adaptive evolution and long-term survival experiments with subseafloor microbes to characterize molecular genetic signatures associated with particular phenotypes.

Field-based research, associated sample analyses, laboratory experiments, and modeling efforts will continue to focus on several key sites. Of particular note are a few field expeditions: IODP Expedition 366 to the Mariana Convergent Margin and the South Chamorro Seamount will recover material and deploy casing within three serpentinite mud volcanoes. This expedition will provide the infrastructure for future sampling and experimental needs within a natural setting that supports an active microbial community in the presence of dissolved hydrogen, methane and sulfate and at a pH of up to ~12.5. This will be a site of ongoing research with international collaborators for years to come. In October 2017, we will also return to North Pond to recover downhole samplers, sensors, and experiments (some of which are part of the instrument strings deployed in 2011 with hundreds of mineral colonization experiments). In addition, we will be collecting large volumes of fluids from various subsurface horizons for assessing fluid microbial activity, genomic potential, and microbe-virus interactions, and we will attempt cross-hole tracer experiments to examine fluid flow pathways. This culminates a multi-year effort of sampling and discovery at North Pond. In May of 2017, the Orphan lab will partake in an Alvin cruise to the Costa Rica Margin to characterize microbial community structure and activity associated with active methane venting. Lastly, a cruise is planned with the Schmidt Ocean Institute for 2018 to a new hydrothermal vent site in the Gulf of California. Stable isotope probing and BONCAT experiments are planned to quantify microbial metabolic activity associated with solid substrates (carbonates), sediments, and fluids.

# III. EDUCATION

## 1. Overall Education Goals and Objectives

The main goal of C-DEBI’s education program is to generate distinctive and targeted activities in and around ocean sciences, in general, and the marine subsurface biosphere, specifically. To achieve this goal, we focus our efforts on three target groups: undergraduate students, graduate students and postdoctoral scholars, and the general public. It should be noted that central to our mission is the integration of our education and research programs—we do not see these as independent pursuits. To engage and retain young people in STEM fields and to develop the next generation of specialists, C-DEBI provides training, mentoring, and professional development opportunities, but we also leverage numerous educational partnerships nationally to work with K-12 students and educators to ensure engagement at all levels.

**2. Undergraduate Students**

As part of our undergraduate education program, we provide several hands-on research activities for community college students and their instructors, including the Community College Research Internship for Scientific Engagement (CC-RISE), the Community College Cultivation Cohort (C4), and community college instructor workshops. For undergraduate students from underrepresented minority (URM) groups at USC, we support the Genomics and Geobiology Undergraduate Research Experience (GGURE), and for undergraduate students across the country who are interested in microbial ecology and biological oceanography, we offer our Global Environmental Microbiology (GEM) course.

CC-RISE is a non-residential REU-style program at two of our partner institutions, the University of California at Santa Cruz (UCSC) and the Marine Biological Laboratory (MBL) in Woods Hole, Massachusetts. It is led by C-DEBI’s Education, Outreach, & Diversity (EOD) Managing Director Stephanie Schroeder, with local oversight at UCSC by Adina Paytan, and at MBL by Julie Huber and Gretta Serres. Twelve academically competitive students (8 at UCSC, 4 at MBL) spent eight weeks during the summer in state-of-the-art research labs. For 40+ hours a week, they carried out experiments and analyses, and participated in professional development seminars that ranged from how to read/write a scientific paper to how to apply to graduate school. Five of the students transferred to 4-year universities in the fall (UC Santa Barbara, UC Davis, UC Berkeley, Bridgewater State University, and the University of Massachusetts).

In previous years, CC-RISE was also offered at USC, but in 2016, with support from the NSF REU program, we converted it into the residential C4 REU program. Eight students (four from California and four recruited at a national level) worked in pairs to characterize three novel bacteria isolated from subseafloor environments. The students learned aerobic and anaerobic culturing techniques, fluorescence microscopy, and some analytical chemistry, and they participated in a 1-week bioinformatics module led by C-DEBI postdoctoral scholar Ben Tully. The students also participated in weekly professional development activities and networking lunches coordinated by Stephanie Schroeder, they interacted with members of their host labs, and they learned about different pathways into science. C-DEBI Director Jan Amend conceived C4 and also hosted four of the students; the other four students worked in the labs of Professors Steve Finkel and Ken Nealson. C-DEBI postdoctoral scholar Ileana Perez-Rodriguez oversaw the day-to-day research activities. In the post-program surveys ([Appendix C](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-C-CC-RISE-and-C4-Student-Evaluation.pdf)), the C4 (and CC-RISE) students provided substantive and predominantly positive feedback, noting that they felt better prepared to succeed at a 4-year university and that their participation in CC-RISE/C4 expanded their views of career options. We should note that educational supplements to both the C-DEBI and the REU grants were used to expand and enhance the CC-RISE and C4 activities. After conclusion of the program, two students also presented their results at the 2016 SACNAS conference, and another student is presenting at the 2016 AGU virtual poster session. In addition, two students are traveling to the 2017 ASLO meeting as part of their dedicated Minority Program.

To best serve the community college students, we also invest resources in the professional development of their instructors. This year Stephanie Schroeder organized a 1-day workshop in April and, in collaboration with the Wrigley Institute for Environmental Studies (WIES), a weekend workshop in November. Some educators wish to learn about deep biosphere research, and others seek accessible activities and lesson plans that help integrate cutting-edge research and new discoveries into their classrooms.  The April workshop included instructors from 10 Los Angeles-area community colleges; the instructors were introduced to current C-DEBI research, and they brainstormed on how to incorporate their new knowledge into current curricula. Postdoctoral researchers Ally Pasulka (Caltech) and Sean Jungbluth (USC) assisted in the process by presenting their research to participants. The November workshop was held on Catalina Island, and instructors from 9 Los Angeles-area schools participated in hands-on activities on a wide range of topics, from marine microbiology to oceanography. Evaluations of both workshops ([Appendix D](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-D-CC-Instructor-Workshop-Evaluations.xlsx)) were positive, calling out in particular the benefits of interdisciplinary topics and networking opportunities.

To stimulate and track teacher application of workshop curriculum to students, two K-16 Educator Small Grants were awarded to CC instructors. The K-16 Educator Small Grant program is a follow-up program for participants of previous C-DEBI K-16 Instructor Workshops (e.g., the CC Instructor Workshop). Los Angeles Trade Technical College instructor, Angela Gee attended the ABLE (Association for Biology Laboratory Education) conference and presented toolkits she developed with C-DEBI and the College of Exploration via a C-DEBI E&O Small Grant. The kits are aligned with specific community college course subjects and were produced through a collaborative process involving scientists, community college faculty, graduate students and education and technology experts. Allesandro Grippo, a Santa Monica College (SMC) instructor, brought 16 SMC students to the Wrigley Marine Science Center for 3 days to gain hands-on training in the field and laboratory. Students enrolled in geoscience courses at SMC were recruited, and workshop content was designed to reinforce and complement geoscience concepts learned in the classroom.

GGURE, a research internship program that targets URMs, builds on a 13-year effort led by Senior Scientist Steven Finkel to recruit and maintain undergraduate students in STEM fields as a part of USC’s Center for Excellence in Genomic Science as part of the National Human Genome Research Institute’s Minority/Diversity Action Plan. There is a part-time program during the academic year and a full-time program over 10 summer weeks; in each case, students carried out microbiological research in a USC lab. In addition, weekly journal clubs provided opportunities for students to delve deeper into research topics while forming a tight research cohort. As reported in external evaluations ([Appendix E](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-E-GGURE-Student-Evaluation.pdf)), these experiences made the students more likely to include research in their career goals.

##### Another flagship component of undergraduate education program is the GEM course, which targets URMs early in their academic careers. Now in its sixth year, this field-based, hands-on, 4-week course is led by USC faculty John Heidelberg and Eric Webb, with directional support from EOD Managing Director Stephanie Schroeder. In 2016, sixteen students participated, many from community colleges across the country. We remain in close contact with all graduates of the course through social media and other means, and we strive to form a community of young researchers with this common experience. More than 80% of the GEM students reported in the external evaluation ([Appendix F](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-F-GEM-Student-Evaluation.pdf)) that the course had a significant impact on their educational goals and careers. The hands-on lab experience and field work were noted as the most meaningful components of the course.

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| **Activity Summary** | **Undergraduate and Community College Programs** |
| Led by | Dr. Stephanie Schroeder, Dr. Jan Amend, Dr. Julie Huber, Dr. Andrew Fisher, Dr. Steven Finkel, Dr. John Heidelberg, Dr. Eric Webb, Dr. Adina Paytan, Dr. Gretta Serres, Dr. Diane Kim, Dr. Ben Tully, Dr. Ileana Perez-Rodriguez, Dr. Sean Jungbluth, Gus Ramirez, Dr. Ally Pasulka, Dr. Angela Gee, Dr. Allesandro Grippo |
| Intended Audience | Undergraduates & community college instructors |
| Approximate Number of Attendees | 250 |

##### ► See more at the [CC-RISE webpage](http://www.darkenergybiosphere.org/education-diversity/for-undergraduates/cc-rise/)

##### ► See more at the [C4 webpage](http://www.darkenergybiosphere.org/education-diversity/for-undergraduates/nsf-c4/)

##### ► See more at the [GEM webpage](http://www.darkenergybiosphere.org/education-diversity/for-undergraduates/gem-course/)

► See more at the [Educator Small Grants webpage](http://www.darkenergybiosphere.org/education-diversity/for-teachers/)

**3. Graduate Students and Postdoctoral Scholars**

The close integration of education and research is particularly evident in our graduate students and postdoctoral scholars. C-DEBI provides both formal and informal training to these early career scientists. First and foremost, graduate students and postdoctoral scholars make up the bulk of the research personnel in the Co-I and Senior Scientist labs. In addition, C-DEBI awards numerous 2-year research fellowships and small grants, hosts an on-line seminar series, and supports a range of professional development opportunities.

At any time, approximately 30-40 graduate students and postdoctoral scholars are working on C-DEBI research in the labs led by Co-I’s Amend, Huber, Fisher, Wheat, and D’Hondt, and the Senior Scientists Orphan, Orcutt, Spormann, Finkel, and Heidelberg. In 2016, these graduate students and postdoctoral scholars received a wide variety of professional training that intertwined research with education and outreach, including the participation in research and training cruises, oral and poster presentations at national and international meetings, invitations to focused workshops, opportunities to deliver classroom and public lectures, inclusion in and leadership of grant proposals, and mentoring of undergraduate and graduate student research.

C-DEBI also invests in the next generation of subseafloor researchers via its fellowship program, funding 10-15 graduate students and postdoctoral scholars each year. These fellows have always integrated education and outreach with their research activities, but as of this year, we require a formal broader impacts statement in the proposal process. The cohort of C-DEBI fellows (together with the C-DEBI graduate students and postdoctoral scholars in Co-I and Senior Scientist labs) constitutes a private forum to discuss research problems, professional development, and future employment opportunities. EOD Managing Director Stephanie Schroeder also sends weekly emails with information on a variety of topics from organizations including, but not limited to, the AGU, National Postdoc Association, Council of Graduate Schools, and National Association of Geoscience Teachers.

The C-DEBI Networked Speaker Series is another opportunity for C-DEBI graduate students and postdocs to interact with the larger community. Speakers are nominated by the community and selected by ExCom. The speakers give live, 30-minute web seminars, followed by a Q&A session. The seminars are recorded for those unable to attend, and C-DEBI hosts ~3/year. In 2016, the speakers were postdoctoral scholars: Katrina Twing (University of Utah) presenting the “Microbial diversity and metabolic potential of the serpentinite subsurface environment”; Olivia Nigro (University of Hawaii) speaking on “Viruses in the oceanic basement: An integral component of microbial life”, and Stephanie Carr (Bigelow Laboratory for Ocean Sciences) asking “Methanosaeta, are you my methanogen?”.

At our annual meeting, C-DEBI partnered with the Metcalf Institute for Marine & Environmental Reporting (University of Rhode Island, Graduate School of Oceanography) through a C-DEBI Education and Outreach Small Grant to engage graduate students and postdoctoral scholars in an all-day professional development workshop, *Building Leadership in Science Communication*. The goal of this workshop was to train these junior scientists in effective science communication to a wide variety of audiences. Participants worked with media experts to develop clear messages via op-ed pieces and to practice speaking with journalists, in addition to learning best practices for using visual media. In the evaluation, participants commented that they valued the hands-on approach of the workshop and feel better equipped to address the media and a variety of different audiences.

Lastly, individual C-DEBI graduate student and postdoctoral scholars participated in a range of professional development activities. For example, USC graduate students Lily Momper and Laura Zinke received C-DEBI travel exchange grants to attend the Deep Carbon Observatory week-long summer school; Montana State University graduate student Alexander Michaud presented his research during the Montana Tech Chemistry & Biochemistry Seminar Series; WHOI postdoc Carolyn Buchwald spoke to middle school students about being a chemical oceanographer; URI graduate student Mary Dzaugis worked with educators to develop curricula and led hands-on investigations for K-8 students to learn about STEM programs; and C-DEBI postdoc Ben Tully organized and ran two bioinformatics workshops, one in conjunction with AGU and the other through the ECOGEO Research Coordination Network. We also continued our partnership with the Agouron Institute in the International GeoBiology summer course, one of the top training courses for graduate students. This intense, multidisciplinary summer course explores the coevolution of the Earth and its biosphere, with an emphasis on how microbial processes affect the environment and leave imprints on the rock record, and included deep biosphere-specific content in lectures by C-DEBI faculty. In the evaluations ([Appendix G](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-G-GeoBio-Student-Evaluation.pdf)), students listed the collaboration and interdisciplinary nature of the course as the aspects that would most likely impact their careers.

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| **Activity Summary** | **Professional Development** |
| Led by | Dr. Stephanie Schroeder, Dr. Jan Amend, Dr. Julie Huber, Dr. Andrew Fisher, Dr. Geoff Wheat, Dr. Steven Finkel, Dr. Victoria Orphan, Dr. Beth Orcutt, Dr. Alfred Spormann, Dr. Steven D’Hondt, Dr. John Heidelberg, Dr. Sunshine Menezes, Dr. Katie Pratt, Dr. Katrina Twing, Dr. Olivia Nigro, Dr. Stephanie Carr, Alexander Michaud, Dr. Lily Momper, Laura Zinke, Dr. Benjamin Tully, Dr. Carolyn Buchwald, Mary Dzaugis, Dr. John Spear, Dr. Doug LaRowe, Dr. Eric Boyd, Dr. Fredrick Colwell |
| Intended Audience | Graduate students, postdoctoral researchers |
| Approximate Number of Attendees | 764 |

► See more at the [Networked Speaker Seminar Series webpage](http://www.darkenergybiosphere.org/outputs-resources/networked-speaker-series/)

► See more at the [Metcalf Institute Professional Development Workshop webpage](http://metcalfinstitute.org/training/scicomm-cdebi/)

##### ► See more at the [GeoBiology Course webpage](https://dornsife.usc.edu/wrigley/geobiology/)

**4. K-12 and the General Public**

C-DEBI, in partnership with institutions across the country, engaged K-12 students in a variety of activities to increase their knowledge about ocean science and subseafloor biosphere research. For example, in collaboration with the USC Wrigley Institute and USC Sea Grant, we sponsored the High School Marine Science Camp for a fifth year. This 1-week camp is a hands-on, inquiry-based program for 20 diverse high school students recruited nationally. C-DEBI also again supported the USC Young Researchers Program, a 10-week internship for 8 local high school students to carry out research with graduate student mentors, culminating with a poster presentation. Six of these students also presented their research at the Great Minds in STEM conference, which is geared towards increasing Hispanic and Latino representation in STEM fields. Based on the C-DEBI eBook *Where Wild Microbes Grow* (created by an Education and Outreach Small Grant), Dieuwertie Kast developed curricula on subseafloor microbiology and brought the material to K-3 students in the Los Angeles area through a K-16 Educator Small Grant. Co-I Geoff Wheat again ran the Seafloor Science and ROV Summer Camp for 6th-8th graders, which emphasizes crucial technology to conduct subseafloor research; community college students were recruited to serve as interns in the camp.

C-DEBI’s general outreach activities range from interactive programs that involve a wide audience to promoting C-DEBI through popular media. This ranges from presentations to middle and high school students, general audience research seminars, cruise blogs, and interviews with the media. C-DEBI also funded Education and Outreach Small Grants to extend our reach. For example, Christopher Petrone of the Delaware Sea Grant, produced nineteen 15-second science videos focused on topics such as *What exactly lies below the seafloor? More than you think!* and *What types of science do researchers use to study the deep biosphere?* Ten of these videos are also available in Spanish and feature C-DEBI postdoctoral fellow Rosa Leon Zayas. Each video has reached, on average, nearly 300 views.

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| **Activity Summary** | **K-12 Programs and General Outreach** |
| Led by | Dr. Stephanie Schroeder, Dr. Geoff Wheat, Dr. Andrew Fisher, Dr. Victoria Orphan, Linda Chilton, Dieuwertie Kast, Dr. David Case, Christopher Petrone, Dr. Rosa Leon Zayas, Erin McParland |
| Intended Audience | K-12 students, general audience |
| Approximate Number of Attendees | 1085 |

# ► See more at the [High School Marine Science Camp](http://dornsife.usc.edu/uscseagrant/summer-science-programs/) webpage

# ► See more at the [Young Researchers Program webpage](http://youngresearchers.usc.edu/)

# ► See more at the [Seafloor Science and ROV Summer Camp webpage](http://www.ssrovcamp.org)

# ► See more at the [Delaware SeaGrant – Project VIDEO webpage](http://www.deseagrant.org/projectvideo)

**5. Performance with Respect to the Strategic Implementation Plan**

**Goal:** *To bring C-DEBI research and the role of subseafloor microbes to the forefront by: 1) increasing microbiology literacy in the general public and at the K-12 levels; 2) engaging and retaining students in STEM fields; and 3) training the next generation of subseafloor researchers*. In our K-12 and general public activities, we rely heavily on partnerships with established organizations for whom these are the core target audiences. At the undergraduate level, we focus heavily on community college students and students from underrepresented minorities and marginalized groups, because we see a myriad of opportunities for some of the highest educational impact. Our most established education entities targets graduate students and beyond; here, our vision is to provide training in state-of-the-art technologies and instrumentation, together with mentoring in science communication, proposal preparation, project management, and other aspects of professional development.

Our *core* *objectives* in K-12 education/general public are to:

* 1. introduce C-DEBI content (e.g., the subseafloor biosphere, extreme microbiology, science and technology) into K-12 classrooms through professional development workshops for educators;
  2. provide hands-on science opportunities for students to engage them in microbiology and oceanography; and
  3. engage the general public in discovery science using public seminars, outreach activities, and social media.

Our *core* *objectives* in undergraduate education are to:

* 1. attract early and potentially undecided undergraduate students into STEM majors and strengthen their interest and passion for science and research; and
  2. provide cutting-edge university research opportunities, especially for community college students and members of underrepresented minorities.

Our *core* *objectives* in graduate and postdoctoral education are to:

* 1. train and nurture the next generation of subseafloor researchers; and
  2. provide professional development opportunities to allow them to expand their transferable skills.

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| **Objective** | **Metric** | **Status/Problems** |
| 1.2 | Develop and conduct 10 varied activities and programs for K-12 classrooms, e.g., high school class visits to USC/other universities, ROV activities partnered with the Wrigley Institute for Environmental Studies, SeaGrant Summer Marine Camp, and guest speakers/lecturers in classrooms or special events | Met |
| 1.1 | Award 2 E&O small grants and 3 K-16 teacher grants to fund the development of educational opportunities and materials on marine deep biosphere topics and to support K-16 teachers who have attended a C-DEBI teacher training program and have incorporated C-DEBI content into their classrooms, respectively | Met |
| 1.1, 3.2 | Create collaborations between C-DEBI science participants and teachers in 6 professional development activities such as C-DEBI community college instructor workshops and partnered programs (e.g., EARTH teacher workshop with MBARI) | Met |
| 1.3 | Increase the number of individuals engaged in each of the three associated categories: contacts (500 in aggregate), members (receive newsletter; 100 in aggregate), and participants (funded in some way; 50 in aggregate) | Met |
| 1.1, 1.3 | Present at 5 informal science events or national education conferences | Met |
| 1.3 | Communicate the deep biosphere in 5 general audience, non-scientific publications | Met |

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| 2.1, 2.2 | Incorporate deep biosphere content in 3 C-DEBI and partner post-secondary programs (e.g., Global Environmental Microbiology (GEM) summer course, Community College Research Internship for Scientific Engagement (CC-RISE), Community College Cultivation Cohort (C4) REU, and the Agouron Institute International GeoBiology Course) | Met |
| 3.1 | Award 10 individuals from varied institutions C-DEBI funding ranging from small research grants, research and travel exchanges, postdoctoral and graduate student fellowships, and E&O small grants | Met |
| 3.1, 3.2 | Support regular and varied methods for professional development exclusive to graduate students and postdoctoral researchers including weekly mailing list postings and an annual retreat | Met |
| 1.1, 1.2, 2.1, 2.2, 3.1, 3.2 | Assess measurable outcomes of program effectiveness using formative and summative evaluations of Very Good or Excellent (4 or 5 out of 5) conducted internally by C-DEBI education staff and by an external evaluator for all programs | Met (CC-RISE, GEM, GGURE, CC Instructor Workshops, etc.) |

**6. Plans for the Next Reporting Period**

C-DEBI is committed to the continued development of broad-based, targeted education programs that train and foster the next generation of deep subseafloor biosphere researchers and on a broader scale, engage and retain STEM researchers.  Our future objectives are to:

1. Create ongoing outreach programs that include standards-based lesson plans and activities delivered to teachers, while providing outreach opportunities for graduate students and postdoctoral researchers.

2. Strengthen partnerships with community colleges by providing cutting edge research to faculty, promoting undergraduate courses and expanding summer research internship programs.

3. Expand the web site to include downloadable lesson plans and activities for teachers using existing partnerships and evolving new ways to enhance existing curriculum.

4. Use networking, existing organizations, social networking tools and local contacts to increase the scope of C-DEBI’s impact.

5. Promote calls for graduate student fellowships, travel grants, and postdoctoral researchers to attract the next generation of innovative scientists.

# IV. DATA MANAGEMENT AND KNOWLEDGE TRANSFER

## 1. Overall Data Management and Knowledge Transfer Goals and Objectives

C-DEBI facilitates the exchange of knowledge, expertise, intellectual and physical resources, experimental methods, and application of new technologies within its diverse community and between the STC and the community at large. This commitment is demonstrated through open access of all new discoveries, sensors, samplers, data, methods, and platforms. This access has many avenues including, but not limited to, the distribution of information through teleconferences, our website, workshops, meetings, newsletters, presentations, technical documents, peer-reviewed publications, educational activities, and outreach events. C-DEBI is also committed to mentoring students and scientists of all ages, including the exchange of personnel among laboratories and professional development.

The overarching objective of Data Management and Integration (DMI) and Knowledge Transfer (KT) is to disseminate C-DEBI scientific discoveries and technical advances both to the scientific community and broader population. This objective has not changed during the transitions from growth in Phase 1 to nurturing in Phase 2. As such, our DMI and KT goals include (1) implementing effective mechanisms to facilitate intellectual exchanges between institutions of various types, (2) maintaining worldwide access to C-DEBI data and information, (3) nurturing a new generation of C-DEBI researchers, (4) developing and making available targeted education, public outreach, and community interactions, and (5) promoting economic growth through technology development. Significant accomplishments and focused objectives of Center activities during the period of performance have been the implementation of an effective data portal, a concentrated effort that continues to archive C-DEBI produced data within the construct of BCO-DMO, the development of an internet-based system for distributing laboratory and analytical protocols, and continued development and implementation of a center-wide bioinformatics program.

## 2. Knowledge Transfer Activities and Organizations

C-DEBI knowledge transfer (KT) occurs on a near-constant basis with numerous organizations, most significantly with those highlighted in [External Partnerships Section V](#_V._EXTERNAL_PARTNERSHIPS_5). Here, we specifically call out KT activities that occurred during the reporting period and focused on overall Goals 1, 2, and 5 above (implementing effective mechanisms to facilitate intellectual exchanges, maintaining worldwide access to C-DEBI data and information, and economic growth and technical development). Other forms of KT (e.g., those involving Goals 3 and 4) are covered in other sections of this report, such as classroom lectures, public presentations, the Network Speaker Series, fellowship and travel grants, professional development, workshops, conferences, field trips, GEM, CC-RISE, C4, GGURE and teachers-at-sea).

One of the most effective tools for transferring knowledge to the scientific community is the publication of peer-reviewed papers. During the reporting period, we published 39 peer-reviewed journal articles, two PhD theses, a data report and a magazine article. Of the 39 articles, 26 included graduate students, postdoctoral fellows, or both. Each of these contributions is posted on our web page and introduced to the community in a monthly newsletter that reaches ~1000 individuals globally. In addition, during the performance period, 112 presentations/posters were given at numerous special sessions and workshops of large national and international meetings hosted by scientific organizations and partners (e.g., AGU, ISME, ASM, ISSM). Other, smaller C-DEBI leadership-hosted workshops and meetings also contributed to knowledge transfer. These meetings included one workshop (18 people) that focused on the “Origins and Movement” of microbes and the C-DEBI Annual Meeting (~80 people) that provides opportunities for experienced and new C-DEBI members to report and discuss recent results, and plan for ongoing and future work.

Several new technological advances were also made during the reporting period. These advances, which can take multiple years for development and implementation, fall within the categories of patent, software and platform developments. New developments include: (1) U.S. Patent No. 62/343,463: Methods for Isolating Nucleic Acids from Samples developed from Co-PI D’Hondt’s collaboration with ExxonMobil Upstream to build on our studies of microbial diversity in deep subseafloor sediment and its relationship to microbial diversity in the surface world (by [Aaron Regberg](https://www.linkedin.com/in/aaron-regberg), Zarath M Summers, A Lucie N'Guessan, John Kirkpatrick and Steven D'Hondt, Provisional Application filed 31-May-2016); (2) a software tool to aid in the binning and reconstruction of microbial genomes from metagenomic data; (3) clustering of metagenomes to produce putative metagenome assembled genomes (MAGs); (4) a small scale ROV for shallow-water science operations and educational activities; (5) a portable autonomous surface vessel for Shallow-Water science operations that include bathymetric mapping and chirp subbotom sonar to assess sediment and fault structures; (6) a retrievable autonomous CTD with the capability to collect samples during ascent based on sensor data collected during descent; and (7) a data portal for C-DEBI publications, data and technical documents. It is anticipated that some of these developments will expand beyond the C-DEBI community.

## 3. Data Management and Integration Activities and Organizations

# The C-DEBI Data Management and Integration (DMI) team is in place to ensure that C-DEBI data and products are archived, shared, and accessible for the long term. The data types and products covered by C-DEBI include a wide variety of geophysical, geological, geochemical, and biological information, in addition to education and outreach materials, technical documents, and samples. The overall DMI goal is to make sure that all data and information generated by C-DEBI-supported researchers as part of their C-DEBI projects are made publically available either following publication or within two years of data generation (see details in our [Data Management Plan](http://www.darkenergybiosphere.org/internal/docs/C-DEBIDataManagementPlan_2015.pdf)). A second goal is to make certain that no C-DEBI researcher is limited by computational resources (e.g., computers or tools). As part of Phase 2, the DMI team also takes the responsibility to make sure the C-DEBI data are integrated in ways to allow larger, more comprehensive analysis.

**a. Making Data Publically Available**

C-DEBI produces many kinds of products that need long term archiving. These include diverse data sets (biological, chemical, physical, and geological), samples, peer-reviewed publications, technological advances with associated engineering drawings and software, educational/outreach materials (such as K-12 and community college lesson plans relating to subseafloor science), and model parameters (e.g., inputs, grids, reaction rates). Biological products include, but are not limited to, molecular data, activity data (isotope abundance, community enzymatic, etc.), frozen samples, living microbial strains, and post-processed molecular data (e.g., 16S rDNA and 16S rRNA sequences, single-cell genome, metagenome and metatranscriptome sequences). Non-biological data will include multi-beam maps, seismic reflection profiles, and thermal, chemical, and physical data from recovered samples of fluids, sediment, rocks, and experiments. Measurements also will be made *in situ* using borehole observatories, drilling platforms, cabled observatories, and coring facilities.

In 2016, we continued our efforts to ensure that all post-embargo data were deposited in appropriate internationally accessible data repositories. The principal repository is the Biological and Chemical Oceanography Data Management Office (BCO-DMO). The C-DEBI DMI team has worked with BCO-DMO to make this location either the primary host of C-DEBI data or to have them provide stable links to data housed in other repositories; with linking to NCBI already available and soon similar links will be available to IODPdb and PRIDE. Products for which a suitable national repository does not exist, such as educational materials, outreach materials, and technical advances, have been posted directly on the C-DEBI main page under the headers 'For Teachers', 'For Everyone', etc. To the extent possible, all such products have also been described in the peer-reviewed literature to ensure public dissemination and long-term accessibility beyond C-DEBI. All C-DEBI intellectual products (publications, technical advances, software, education and outreach materials) are directly linkable by searching the main C-DEBI page. This provides direct electronic access to the data repository, publication, and protocols.

We have made major progress in our mandate to deposit C-DEBI data in public repositories. Over the last year, the projects on the [C-DEBI BCO-DMO webpage](http://www.bco-dmo.org/program/554979) have increased from 17 to 50. There are several others queued in the BCO-DMO quality check phase. The majority of the Small Grants programs that ended more than two years ago have deposited their required data in public repositories. Those who have not yet complied have been reminded to do so as soon as possible, and many have been in contact with BCO-DMO to complete this task. Of the 97 completed research grant projects (small grants, fellowships and exchanges), most have completed generating data (40%) or are non-data-generating (10%), while the rest are in the process of generating data (26%) or are of an unknown status (24%). Currently, project compliance of having data deposited in a public repository is greater than 77%. In the non-compliance cases, all PIs have been made aware of the requirement to deposit ultimately to BCO-DMO, and are currently unable to receive any additional C-DEBI resources until they are known to be in compliance.

**b. Providing Computational Resources to C-DEBI Researchers**

In 2016, C-DEBI continued to maintain a computing resource to accommodate data analysis on scales too large for laboratory computational resources, but too small (or poorly designed) for high powered computing centers. Currently, there are 13 C-DEBI researchers with access, provided on a rotating basis. These users are members of the larger C-DEBI community, and include graduate students, postdoctoral researchers and faculty from multiple labs and grants, not just the Co-Is and Senior Scientists. The need for this service continues to grow, and we expect continued use in future years.

Beyond basic access to a maintained computer resource, several initiatives have been implemented to make sure C-DEBI researchers are not limited by any step in the bioinformatics process. One important aspect of this is the training of researchers on available tools. To this end, prior to the American Geophysical Union (AGU) Fall Meeting in December 2016, Dr. Benjamin Tully and graduate student Elaina Graham hosted a second annual bioinformatics training session at Stanford University for 12 C-DEBI researchers. The attendees were also from the broader community, including 5 faculty, 2 post docs, 4 graduate students, and 1 lab tech. The faculty were from Dartmouth, University of Texas El Paso, Hamilton College, University of South Carolina, RPI, UC Santa Cruz, UC Santa Barbara, and Stanford. Information from that training session will be translated into various resources for general use by the scientific community, including video tutorials and aggregated digital protocols at Protocols.io.

**c. Expanding the Impact of C-DEBI Data through External Partnerships and Collaborations**

To visualize and query all C-DEBI data in new ways, we provide and support several existing and proposed data platforms. We also now request that C-DEBI researchers put both successful and unsuccessful protocols into the [C-DEBI group on Protocols.io](https://www.protocols.io/groups/center-for-dark-energy-biosphere-investigations).

We have also formally accepted an offer by Drs. DeLong, Hurwitz, and Wood-Charlson for C-DEBI to collaborate with their EarthCube Building Block proposal, “Planet Microbe.” C-DEBI believes collaborations are the soundest way to meet our goals for data discovery, integration, synthesis, and open sharing, and we strive to leverage available infrastructure and to partner with excellent groups like “Planet Microbe.” We are excited to collaborate at all stages of Planet Microbe, from development and validation to implementation and sustainability. Dr. John Heidelberg, Associate Professor of Biology at USC and the C-DEBI Data Management and Integration manager, and Dr. Benjamin Tully, Bioinformatics Specialist for C-DEBI, can supply the expertise and support needed to validate current C-DEBI data. In addition, they will be instrumental in the development of standardized workflows to ensure that future C-DEBI data will also be contributed to and validated in Planet Microbe. While this Building Block has not yet been funded, we will continue to work with the team to develop meaningful data intermigration sites for C-DEBI data. For example, Dr. Hurwitz was invited to present on iMicrobe at the C-DEBI annual meeting in October.

Finally, we are continuing our efforts to make C-DEBI data available to the larger ocean sciences community. To this end, C-DEBI has worked to maintain a strong presence in the larger EarthCube community. Dr. Heidelberg is a member of the steering committee for the ECO-GEO RCN and, together with PI Amend and other C-DEBI members, participated in the EarthCube all-hands meeting in 2016, and Dr. Tully co-ran the [ECOGEO Workshop: Introduction to Environmental 'Omics](https://www.earthcube.org/event/ecogeo-workshop-introduction-environmental-omics) ([training modules on protocols.io](https://www.protocols.io/researchers/elisha-woodcharlson/protocols)).

## 4. Performance with Respect to the Strategic Implementation Plan

Our data management and knowledge transfer goal is to implement effective mechanisms and pathways to facilitate the exchange and application of knowledge, expertise, physical resources, and novel methods and technologies within the STC and between the STC and the broader community. The overall data management plan is in place to 1) assure all data generated from the STC are deposited in publically accessible data repositories, 2) efficiently allow STC researchers tools and computational resources that allow them to efficiently perform data analysis, and 3) develop and maintain a data portal for visualization and hypothesis generation from the STC data.

**Target 1:** Innovations are imported/exported/shared and partnerships are developed with other fields, research institutions, industry and government

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| **Metric** | **Status/Problems** |
| Publish and promote scholarly activity via 10 publications | Met |
| Continue to develop research collaborations through networking at 2-3 interdisciplinary meetings and talks/posters/exhibition at 2-3 conferences | Met |
| Lead 3 C-DEBI-focused meetings or special sessions at national or international meetings | Met |
| Enhance, develop, or commercialize tools, analytical capabilities, software products, sensors and platforms (2 per year) | Met |

**Target 2:** New innovation in the field is communicated through web tools, publications, media, presentations, and educating the next generation of researchers and ocean stewards.

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| **Metric** | **Status/Problems** |
| Continue to ensure all data generated through the STC are in stable public data repositories within 2 years of generation (or for graduate students upon defense of the dissertation or thesis) | Met (details above) |
| Continue to develop web-based data portal bringing data together from various repositories for synthesis efforts | Met |
| Train researchers in new tools for data analysis by producing 3 webinars on data analysis tools per year and 2 small workshops for data analysis | Met |
| Engage 20 new undergraduates to the fields of microbiology and oceanography and mentor 10 graduate students in C-DEBI fields | Met |
| Communicate with the public through non-scientific journals via social and journalistic media (5 significant contributions) | Met |

## 5. Plans for the Next Reporting Period

C-DEBI is continuing with its vision for long-term data management and knowledge transfer activities for the next reporting period, consistent with the current practices and those outlined in the renewal proposal and in response to feedback from previous NSF Site Review committee members and NSF personnel.

# V. EXTERNAL PARTNERSHIPS

## 1. Overall External Partnerships Goals and Objectives

C-DEBI supports cross-disciplinary and cross-institutional partnerships that facilitate, augment, and expand the education, training, and research opportunities of Center participants.

## 2. Activities Conducted as Part of Partnerships

In Phase 2 of C-DEBI, we continue our strong, long-standing relationships with several high-profile external partnerships, both in research and education efforts. Of particular note on the research side are partnerships with the International Ocean Discovery Program (IODP), University-National Oceanographic Laboratory System (UNOLS), National Deep Submergence Facility (NDSF), Japan Agency for Marine-Earth Science and Technology (JAMSTEC), the Sloan-Foundation funded Deep Carbon Observatory (DCO), US Department of Energy (DOE), International Continental Drilling Program (ICDP), Schmidt Ocean Institute (SOI), NASA Astrobiology Institute (NAI), Los Alamos National Laboratory (LANL), and ExxonMobil. On the education side, we again partnered with the Agouron Institute to train graduate students in geobiology, and we continue to leverage our partnerships with the USC Wrigley Institute for Environmental Studies and USC SeaGrant programs on our undergraduate and high school experiences. We again offered our CC-RISE program at UCSC and MBL, and we used NSF-REU support (including a supplement) to train community college students in microbial culturing and other C-DEBI research.

**IODP**

Since its inception, C-DEBI science has been integrally connected to IODP's focus on exploring and documenting the deep biosphere. During Phase 1, C-DEBI scientists led several expeditions (Expeditions 327, 329, 336) and were integral science party members of many others (Expeditions 323, 330, 331, 337, 339, 347, and 353). Research continues from those expeditions in Phase 2 and the current reporting year; many papers from these expeditions were included in a recent special issue of *Frontiers in Microbiology*. Bracketing the start and end of the current reporting period, C-DEBI scientists led IODP Expeditions 357 (Atlantis Massif; Senior Scientist Orcutt) and 366 (Mariana Convergent Margin; Co-PI Wheat), both of which have deep biosphere science as a lead component. Moreover, C-DEBI scientists participated in several other expeditions throughout the year: 360 (SW Indian Ridge Lower Crust/Moho), 364 (Chicxulub Impact Crater), 365 (NanTroSEIZE Shallow Megasplay Long-term Borehole Monitoring System) and 370 (T-Limit of the Deep Biosphere off Muroto). IODP has also benefitted C-DEBI efforts through salary, research, workshop, and travel support for scientists, educators, engineers, and students, both within the US and internationally.

**UNOLS and NDSF**

C-DEBI relies heavily on the UNOLS fleet of research vessels for expeditions, as well as the NDSF fleet of remotely operated vehicles (e.g., *Jason*), autonomous underwater vehicles (e.g., *Sentry*), and the human occupied vehicle *Alvin*, particularly for CORK servicing activities and coring expeditions. Co-PI’s Amend and Huber, among other C-DEBI members, attended the DESCEND-2 (2nd DEep Submergence science for the Next Decade) workshop last January, focused on defining the critical scientific goals of the submergence community and the technological directions that will be required to address these goals.

**JAMSTEC**

C-DEBI collaborates closely with the Geomicrobiology Group (led by Fumio Inagaki) at the JAMSTEC-Kochi Institute for Core Sample Research. In the past year, several C-DEBI scientists from IODP Expedition 357 (Atlantis Massif) utilized state-of-the-art facilities at JAMSTEC for processing samples, and C-DEBI scientists from IODP Expedition 370 (T-limit of the deep biosphere) were based at these facilities throughout the expedition. Members of C-DEBI and JAMSTEC have also partnered on other international expeditions and proposals for new expeditions.

**DCO**

The Sloan Foundation-funded DCO is organized into four research communities, including one on ‘deep life’, which is dedicated to assessing the nature and extent of the deep microbial and viral biosphere. This community, co-chaired by Mitch Sogin and Kai-Uwe Hinrichs, funds scientific networking opportunities (e.g., workshops), instrumentation, infrastructure, and focused research initiatives (e.g., the Census of Deep Life (CoDL) and a project on rock-hosted communities). C-DEBI member Rick Colwell is the lead proponent of the CoDL and a member of the DCO Deep Life Steering Committee. C-DEBI Co-PI D’Hondt is also a member of the DCO Deep Life Steering Committee. Several C-DEBI samples have been included in the CoDL sequencing efforts and others are in the queue. C-DEBI supported scientist Karen Lloyd is a member of the DCO Executive Committee. Senior Scientist Orcutt and C-DEBI member Karyn Rogers serve on the Task Force 2020 to chart the future of the DCO program.

A joint DCO/C-DEBI workshop on Origins and Movements of Subsurface Microorganisms was held May 9-10 at USC organized by Tom Kieft and Doug LaRowe. It addressed fundamental questions about how deeply-dwelling microbial species become adapted to diverse subsurface conditions and become geographically distributed in what appear to be hydrogeologically isolated settings. Experts in groundwater dating, hydrogeological flow and transport modeling, genomics, and evolutionary microbiology were brought together to present plenary lectures and participate in breakout sessions to discuss the current state of knowledge as well as opportunities for new field and laboratory studies.

**ICDP**

The ICDP is supporting several deep life projects, including drilling of the Oman ophiolite and the Death Valley rift zone. C-DEBI is partnering with ICDP by supporting complementary aspects of these projects, with a particular focus on their microbial communities. Both of these ICDP projects also link to the NAI (see below); Duane Moser, Co-I on the ‘Life Underground’ team is playing a central role in the Death Valley drilling plans, and Alexis Templeton, PI on the ‘Rock-Powered Life’ team is directly involved in the Oman drilling activities.

**DOE**

UCSC-based C-DEBI researchers were invited in 2016 to join scientists from the Lamont-Doherty Earth Observatory, Columbia University, and Battelle's Pacific Northwest Laboratory on a proposal to the DOE's Carbon Storage Assurance Facility Enterprise (CarbonSAFE): Integrated CCS Pre-Feasibility. This proposal is for analysis of parts of the Juan de Fuca plate that could potentially be used for large-scale carbon storage; this proposal was recently recommended for funding. Work to take place in 2017-18 will include analysis of single- and cross-hole pressure, temperature, and tracer response to perturbation, as a means to assess hydrogeologic properties of this environment that could impact CO2 injection. This work builds directly from C-DEBI studies associated with IODP Expedition 327, modeling, and related post-drilling expeditions. A formal decision about this DOE project has not yet been made, but is anticipated in the next 4-5 weeks. Assuming a positive decision, more details will be provided in the 2017 Annual Report.

**SOI**

SOI is a private foundation that serves as an oceanographic operator for the seagoing community by providing ship and vehicle time via community solicited, peer-reviewed proposals. A number of ExCom members and C-DEBI investigators have participated in cruises aboard the SOI research vessel *Falkor*. For example, Associate Director Huber headed to sea with SOI in late November 2016 to explore newly discovered hydrothermal vents along the Mariana BackArc. Many C-DEBI members have submitted proposals to SOI for the next round of expeditionary proposal selections and are awaiting decisions on their outcome.

**NAI**

‘Life Underground’ is one of the NAI CAN-6 teams, with 5 year funding through 2017. This cross-disciplinary team, led by PI Amend, is using field, laboratory, and modeling approaches to detect and characterize microbial life in the subsurface—predominantly, but not exclusively, on the continents. C-DEBI and NAI are sharing key personnel, jointly developing down-hole biomass detection capabilities using deep UV microscopy, modeling microbial metabolism potential in marine sediments globally, and coordinating several education and outreach efforts. An NAI Postdoctoral Fellowship was recently awarded to Elizabeth Trembath-Reichert from Victoria Orphan’s lab to work between Huber and Amend on microbial activity in North Pond crustal fluids. As another example, former C-DEBI postdoctoral scholar Annie Rowe (working with Ken Nealson) is now working with Amend and Moh El-Naggar (Physics at USC), funded by the NAI.

**LANL**

C-DEBI (through the UCSC Hydrogeology group) is collaborating with researchers in the Earth and Environmental Sciences Group at LANL to develop complex simulations of seafloor hydrothermal circulation. Co-PI Fisher and graduate student, Esther Adelstein, visited with LANL colleagues in Summer 2016, to collaborate on grid generation and modeling, and postdoctoral researcher, Tess Weathers, is developing reactive transport models using the same numerical tools. Fisher and colleagues have also collaborated with development of geostatistical tools that can be applied to generation of complex permeability fields, and irregular boundaries, for improved representation of the upper ocean crust. Collaborative work with LANL and other colleagues is expected to continue into 2017.

**ExxonMobil Upstream**

C-DEBI (through Co-PI D’Hondt) collaborated with ExxonMobil Upstream to build on our studies of microbial diversity in deep subseafloor sediment and its relationship to microbial diversity in the surface world. A recent product of this collaboration is U.S. Patent No. 62/343,463 (Methods for Isolating Nucleic Acids from Samples) by [Aaron Regberg](https://www.linkedin.com/in/aaron-regberg), Zarath M Summers, A Lucie N'Guessan, John Kirkpatrick and Steven D'Hondt (Provisional Application filed 31-May-2016).

**Education**

The interdisciplinary nature of C-DEBI research lends itself magnificently to a diverse array of external education partnerships as well. One of our primary education goals is to train the next generation of deep subseafloor biosphere researchers, and to do so, we partner with one of the top training courses for graduate students, the Agouron Institute International GeoBiology summer course. In 2016 (and in previous years), this course was co-directed by C-DEBI member Dr. John Spear, Colorado School of Mines, and featured several C-DEBI members as guest lecturers. We also partnered with the Metcalf Institute for Marine & Environmental Reporting for our annual professional development workshop targeting graduate students and postdoctoral researchers. Just as C-DEBI and the Agouron Institute course share key personnel (administrative and instructional), so do C-DEBI and the USC Wrigley Institute. This facilitates our training of undergraduates through programs such as the Global Environmental Microbiology course (based heavily on the successful GeoBiology program) and a growing ROV education program at the Institute’s marine lab on Catalina Island. The facility is also the site of our high school program, run by the USC SeaGrant program, and held at the Wrigley Institute. Our outreach partners have grown to include the Monterey Bay Aquarium Research Institute, the College of Exploration, and community colleges across the country that enable us to train teachers at the K-16 levels.

## 3. Performance with Respect to the Strategic Implementation Plan

Our external partnership goal is to engage and support cross-disciplinary and cross-institutional partnerships that facilitate, augment and expand the education, training and research opportunities of Center participants. Partnerships among individuals, institutes, organizations, and programs are the core of C-DEBI research and educational efforts. One of the strengths of these partnerships is the quality and broad appeal of publications. C-DEBI has a range of cross-disciplinary and cross-institutional collaborations that have transformed our view of subsurface microbial conditions, activity, and mechanisms within the hydrologic and geochemical context of fluid flow within the oceanic crust. Another strength of C-DEBI partnerships is the web of interaction of C-DEBI community within other organizations and programs and the joint efforts of these organization and programs in collaboration with C-DEBI to promote and facilitate synergetic research objectives. These partners also extend to education and diversity efforts, leading to a community of junior scientists that are engaged in cross-disciplinary and cross-institutional training activities and exchanges.

**Target 1:** Strong cross-disciplinary research projects and strong cross-institutional programs are demonstrated in all aspects of Center activities, including publications, presentations, proposals, educational exchanges, and educational programs.

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| --- | --- |
| **Metric** | **Status/Problems** |
| Publish 5 (in aggregate) cross-disciplinary papers per each of the three research themes with support from calls for small research and travel grants to facilitate the interaction of dispersed Center researchers | Met |
| Submit 2 cross-disciplinary and cross-institutional proposals | Met |
| Support 2 interdisciplinary workshops or meetings in concert with other national programs | Met |
| Fund 10 graduate students, postdoctoral fellows and C-DEBI community scientists in their pursuit of generating data or developing cross-discipline techniques and tools to further Center objectives | Met |
| Provide the funds that allow 3 student/researchers the opportunity to participate in research expeditions or travel to another institution to expand the scope of their education/research in the use of novel techniques and tools | Met |

**Target 2:** Partnerships are developed with other fields, research organizations, industry, government, and foundations.

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| **Metric** | **Status/Problems** |
| Build 10 (total) partnerships by networking at interdisciplinary meetings, developing industrial and governmental partners, and targeting partnerships and interactions for new applications of existing or budding technologies | Met |
| Develop documents and materials that highlight significant results through C-DEBI research, education, and diversity programs suited to specific organizations, foundations, and programs to form the basis of a dialogue between C-DEBI and that organization to further fiscal and research needs | Met |

## 4. Plans for the Next Reporting Period

C-DEBI has had long-standing partnerships with most of these external partners and the close collaborations are likely to continue at similar levels of commitment for the foreseeable future.

# VI. DIVERSITY

## 1. Overall Diversity Goals and Objectives

C-DEBI seeks totrain a new, diverse generation of undergraduate, graduate and postdoctoral researchers within an integrated and collaborative multidisciplinary community. We are committed to improving access and support for members of underrepresented and marginalized groups to be able to succeed in STEM fields. It should be noted, however, that the activities intended to enhance diversity do not happen in a vacuum—they are purposefully integrated with C-DEBI’s fundamental research, education, and outreach missions.

## 2. Activities Which Enhance Diversity at the Center

C-DEBI has made gains in gender representation of our research and administrative participants (see diversity statistics below). Women are still underrepresented in the natural sciences, and of the C-DEBI participants who reported their gender, 55% are women. Particularly encouraging are the numbers among undergraduate students (67%), graduate students (59%), and postdoctoral scholars (55%), who represent the future in the field. We also continue to work toward increasing underrepresented minorities in C-DEBI by promoting deep subsurface research through Minority Professional Organizations and national networks. This year, we disseminated program and graduate training opportunities with partners such as the Institute for Broadening Participation (IBP), the Society for Advancing Chicanos and Native Americans in Science (SACNAS), and the broader STC Education and Diversity network.

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| --- | --- | --- |
| **C-DEBI Research and Administrative Participants** | **Women (Reported)** | **Men (Reported)** |
| Faculty (55) | 33% | 67% |
| Other Research Scientist (11) | 25% | 75% |
| Postdoctoral (31) | 52% | 42% |
| Graduate Student (50) | 57% | 41% |
| Undergraduate (82) | 67% | 33% |
| Pre-college (28) | 46% | 54% |
| Teacher / Educator (36) | 50% | 28% |
| Other Participant (4) | 100% | 0% |
| Staff (5) | 80% | 20% |
| **Total (302)** | **53%** | **44%** |

One individual identified as transgender and 11 individuals (4%) did not report gender. One individual identified a disability, 214 individuals (71%) reported none, and 87 individuals (29%) did not provide information.

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| --- | --- | --- | --- | --- | --- | --- |
| **C-DEBI Research and Administrative Participants** | **White (all ethnicities)** | **White, Hispanic/ Latino** | **Native Hawaiian or Other Pacific Islander** | **Asian** | **African American** | **Native American** |
| Faculty (55) | 84% | 4% | 0% | 2% | 0% | 0% |
| Other Research Scientist (11) | 91% | 0% | 0% | 0% | 0% | 0% |
| Postdoctoral (31) | 61% | 13% | 10% | 0% | 0% | 0% |
| Graduate Student (50) | 80% | 10% | 0% | 4% | 0% | 0% |
| Undergraduate (82) | 17% | 27% | 2% | 6% | 40% | 0% |
| Pre-college (28) | 14% | 57% | 0% | 0% | 21% | 0% |
| Teacher / Educator (36) | 36% | 11% | 0% | 17% | 28% | 0% |
| Other Participant (4) | 75% | 0% | 0% | 0% | 25% | 0% |
| Staff (5) | 40% | 20% | 0% | 40% | 0% | 0% |
| **Total (302)** | **50%** | **18%** | **4%** | **6%** | **13%** | **0%** |

The nine individuals (3% of C-DEBI participants) who identified as multiracial or mixed race, specified the following: Asian/mixed race; black/white/other; Cape Verdean/Multiracial; native Hawaiian or other Pacific Islander & white; Hispanic or Latino/multi; African American/multiracial; African American/multiracial; Asian/white. There are 27 individuals (9% of C-DEBI participants) who did not provide information. U.S. Citizens comprised 72% of C-DEBI participants, Permanent Residents 3%, other non-U.S. Citizens 7%, and 18% did not provide information. One U.S. Citizen reported holding dual citizen status with Canada. Of the non-U.S. Citizens, two identified as being from Canada, one from the UK, one from Belgium, one from Iran, one from Argentina, one from Chile, one from Turkey, and 14 individuals did not specify.

C-DEBI continues to build on program successes of the past four years by expanding and evaluating four distinct projects targeting underrepresented minorities, women, and first generation and low-income students: the Global Environmental Microbiology (GEM) course, the Genomics and Geology Undergraduate Research Experience (GGURE), the USC Young Researchers Program, and the High School Marine Science Camp. These programs enrich the scientific skills of students through a combination of field-based research with professional development activities.

## 3. Impact of Programs and Activities on Enhancing Diversity at the Center

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| **Program** | **Number of Participants** | **Diversity Objective** | **Measurement of Outcomes** |
| NSF REU: Community College Cultivation Cohort (C4) | 8  (started in 2016) | NSF REU for community college students recruited at a national level | External evaluation, Longitudinal tracking |
| Community College Research Internship for Scientific Engagement (CC-RISE) | 6-12  (31 in 4 years) | Summer research internship for community college students near UCSC and MBL | External evaluation, Longitudinal tracking |
| Genomics and Geology Undergraduate Research Experience (GGURE) | 28  (academic year) | Academic year and summer research internship program for underrepresented undergraduate students at USC | External evaluation |
| Global Environmental Microbiology (GEM) Summer Course | 16  (95 in 6 years) | Hands-on experience for 2- and 4-year undergraduate students in environmental microbiology | External evaluation,  Retrospective survey,  Longitudinal tracking |
| Young Researchers Program | 5  (27 in 5 years) | Research lab experience for high school students | Summary report |
| Marine Science Camp | 20  (99 in 5 years) | Hands-on exploration of oceanography for high school students | Summary report of survey |

## 4. Performance with Respect to the Strategic Implementation Plan

Our diversity goal is to implement programs that introduce Center research and findings to members of underrepresented groups and strengthen the STEM pipeline. Through its academic programs, C-DEBI promotes inclusion and retention among underrepresented groups, including women and first-generation college students. With a strong base established to increase STEM diversity at all levels, we are developing and exporting distinctive, in-depth education and research experiences that encourage historically underserved students. These initiatives are targeted toward three primary sectors: 1) pre-college; 2) undergraduate and community college populations; and 3) early-career and established scientists. Diversity initiatives are incorporated into all education programming.

Our two main *objectives* are to:

1. increase the diversity in the C-DEBI community, especially underrepresented minorities, of C-DEBI graduate students and post-doctoral scholars; and
2. help minority undergraduate students (in community colleges and at USC), who are interested

in STEM majors, develop a path to a career in a STEM field.

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| **Objective** | **Metric** | **Status/Problems** |
| 1 | Promote C-DEBI research opportunities to diverse audiences through partners (e.g., Institute for Broadening Participation) to increase the diversity of graduate students and postdoctorals across the center | Met |
| 2 | Develop or expand programs to attract underrepresented students into STEM fields | Met |
| 2 | Assess measurable outcomes of program effectiveness using formative and summative evaluations of Very Good or Excellent (4 or 5 out of 5) conducted internally by C-DEBI education staff and by an external evaluator for all programs | Met |
| 2 | Introduce C-DEBI science with appropriate resources and training to 1 institution and/or educator that primarily serves underrepresented groups | Met |

## 5. Plans for the Next Reporting Period

Our future goals are to:

1. Actively encourage undergraduates to progress to graduate school in areas of deep subsurface research by promoting summer research or intensive programs being led at C-DEBI networked institutions
2. Continue to leverage support services and potential connections, organizations and institutional resources within partnering Universities to promote diversity
3. Actively promote all opportunities throughout the Center to underrepresented groups and recruit at all levels of Center activity
4. Inform and encourage the C-DEBI community to participate in conferences and outreach that engages them with underrepresented students to promote recruitment into C-DEBI fields
5. Continue to leverage SACNAS involvement to promote and provide financial support to undergraduate and graduate students from C-DEBI to present research at the annual SACNAS conference
6. Continue support and mentoring for the USC SACNAS Chapter linking them to SACNAS Chapters at C-DEBI partnering institutions and SACNAS leadership.

# VII. MANAGEMENT

**1. Overall Organizational Strategy**

Our management plan facilitates the achievement of the principal scientific, education, and diversity goals of C-DEBI. C-DEBI management is composed of these major leadership groups: Directorship; Executive Committee; Administration; Research; Knowledge Transfer, Data Management and Integration; and Education, Outreach and Diversity. The major advisory groups for C-DEBI are the External Advisory Board; Ethics Panel; External Evaluator; and the Education & Outreach Steering Committee. We maintain a simple hierarchy in the management structure (rectangles below) with several advisory groups (ovals below) to encourage communication and collaboration, as well as provide transparency in decision-making. Their roles and interactions are described below and further detailed in our [Operations Manual](http://www.darkenergybiosphere.org/wp-content/uploads/C-DEBIOperationsManual_2016f.pdf).

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**Directorship**

The Center is led by the Director, PI Jan Amend (USC), the Associate Director, Julie Huber (MBL), and the Managing Director, Rosalynn Sylvan. The Director is responsible for overall C-DEBI coordination and performance. He provides leadership in C-DEBI scientific, education, diversity, outreach, and administrative activities; he represents C-DEBI in interactions with USC administration and funding agencies; and he promotes the Center worldwide.

The Associate Director is the ‘right hand’ of the Director; she assumes all responsibilities and powers of the Director should he, for any reason, be unable to carry out his duties. Together with the Managing Director, she coordinates the grants program and communicates with grant recipients about outcomes, products, and dissemination of results.

The Managing Director manages fiscal matters and grants administration and oversees the administrative staff.

**Executive Committee (ExCom)**

The Executive Committee (ExCom) manages, supports and leads the direction of the Center’s science initiatives. ExCom also provides guidance to integrate research, education, and data across the Center. ExCom coordinates with the Senior Scientists (see Research management section below) on C-DEBI research directions. ExCom generates calls for proposals and serves with the Senior Scientists as the review panel, with mechanisms to avoid conflict-of-interest.

ExCom consists of seven permanent members and two rotating members. The permanent members are Director and PI Jan Amend (USC), Associate Director and co-PI Julie Huber (MBL), co-PI Steven D’Hondt (URI), co-PI Andrew Fisher (UCSC), co-PI C. Geoffrey Wheat (U Alaska-Fairbanks), Data Management Director John Heidelberg (USC), and Education Director Stephanie Schroeder (USC). Heidelberg and Schroeder were added to ExCom to integrate our Data Management and Integration activities and our Education, Outreach, and Diversity programs with C-DEBI research. The rotating members consist of Senior Scientists (see Research management section below) added to complement the research expertise on ExCom and serving 15-month terms (currently Steven Finkel (USC) and Victoria Orphan (CalTech).

ExCom maintains communication via weekly videoconference meetings, two annual face-to-face meetings, and ad hoc meetings at selected C-DEBI, national and international meetings, with participation by the Managing Director and as needed by members of the Administration; Research; Knowledge Transfer, Data Management and Integration; and Education, Outreach and Diversity Teams.

**Administration**

The administrative staff, led by Managing Director Rosalynn Sylvan, is based at USC, where they manage the Center’s day-to-day activities. They link to C-DEBI activities at the partner institutions and communicate with all participants worldwide. The Managing Director manages fiscal matters and grants administration and with the Education Director, Stephanie Schroeder, oversees the administrative staff. The Managing Director attends the weekly ExCom videoconference meeting and any other face-to-face ExCom meetings as the administrative liaison. Since 2015, the Managing Director has been operating remotely at Texas A&M University as a Visiting Scholar at the International Ocean Discovery Program (IODP) with regular email and telecommunications in addition to the weekly administrative and ExCom meetings via videoconferencing.

The Data Manager, Matthew Janicak, is responsible for supporting the database infrastructure (see Data Management and Integration below) and development and maintenance of the website and other community communications. The Administrative Assistant, Nerissa Rivera-Laux, implements day-to-day activities of the center and is responsible for meeting coordination.

**Research**

C-DEBI Phase 1 major field programs are led by members of ExCom, while cross-cutting research themes are led by ExCom and the Senior Scientists. Co-PI Fisher leads the Juan de Fuca Ridge field program, Co-PI D’Hondt leads the South Pacific Gyre field program, and Co-PI Wheat leads the North Pond and Dorado Outcrop field programs. As detailed in the [Research Section II](#_II._RESEARCH_3) above, we have transitioned to three new research themes related to the renewal phase to encourage synthesis and integration across themes and sites. Five Senior Scientists were added to C-DEBI leadership to complement the ExCom expertise on these themes: *Fluxes, Connectivity, and Energy* (Theme 1); *Activities, Communities, and Ecosystems* (Theme 2); and *Metabolism, Survival, and Adaptation* (Theme 3). The Senior Scientists are Steven Finkel (USC), John Heidelberg (USC), Beth Orcutt (Bigelow Laboratory for Ocean Sciences), Victoria Orphan (California Institute of Technology), and Alfred Spormann (Stanford University).

**Knowledge Transfer, Data Management and Integration**

Knowledge Transfer is central to all of C-DEBI’s research, education, and outreach programs, and hence, it is the responsibility of all our senior personnel. The Knowledge Transfer Director, Geoff Wheat, coordinates and tracks the various knowledge transfer activities, with a special focus on dissemination of scientific and technical knowledge, increasing public awareness of the subseafloor biosphere, and promoting development and application of novel technologies through commercialization and entrepreneurial use of C-DEBI products.

The Data Management and Integration (DMI) team has the primary objective to make C-DEBI data and products accessible to the world. The products include C-DEBI publications, data generated by C-DEBI projects, documentation of technological advances, and products for education and outreach. Renewal Senior Scientist, John Heidelberg leads the DMI effort, with support from personnel at USC (Data Manager Matthew Janicak and Bioinformatics Postdoctoral Benjamin Tully) and URI (Data Portal Lead Robert Pockalny). The DMI Director is also responsible for ensuring that C-DEBI participants have access to the Center’s computational resources and/or bioinformatics expertise, as well as making certain C-DEBI generated data are properly deposited in public archives and databases, including future EarthCube initiatives.

**Education, Outreach, and Diversity (EOD) Administration**

The EOD team is based at USC and develops, implements, and coordinates EOD programs and activities. The Education Director, Dr. Stephanie Schroeder, provides oversight, leadership, and commitment to the integration of C-DEBI research with our EOD efforts at all levels. The Education Director also leads the professional development and mentoring efforts for undergraduate and graduate students, postdoctoral scholars, and K-12 teachers, as well as serves as review chair of the small education and outreach grants proposals. The Diversity Director reports to the Education Director and lead programs to entrain members of underrepresented groups into STEM fields with a special focus on microbiology, geochemistry, and oceanography. The Diversity Director also expands the reach of C-DEBI through social media communication. The current Diversity Director, Gwen Noda (CV in [[Appendix H](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-H-Diversity-Director-Noda-CV.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2015/12/Appendix-J-External-Advisory-Board-CVs.pdf)), was hired in October and replaced the previous Diversity Director, Dr. Leticia Sanchez, who resigned from the position.

**External Advisory Board**

The External Advisory Board (EAB) provides an annual assessment of the science, education, mentoring, management, and functioning of C-DEBI to the Directorship. The five member committee of national and international leaders in both science and education includes chair Susan Humphris (WHOI), Doug Bartlett (Scripps), Jon Kaye (Moore Foundation), Rina Roy (American River College), and Judy Wall (University of Missouri). The EAB met with the C-DEBI leadership at the 2016 C-DEBI Annual Meeting to discuss future research and education directions, and reports their recommendations to the directorship confidentially. The chair of the EAB will present their assessment at the Site Visit. Jon Kaye, Program Director of the Marine Microbiology Initiative at the Gordon and Betty Moore Foundation (CV in [[Appendix I](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-I-External-Advisor-Kaye-CV.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2015/12/Appendix-J-External-Advisory-Board-CVs.pdf)), was added to the EAB this year to represent a foundation perspective for Phase 3 of C-DEBI (i.e. the long-term future beyond the STC).

**Ethics Panel**

The Ethics Panel advises ExCom on any issue pertaining to ethics, including concerns regarding administration, funding, and scientific conduct. This Panel handles all C-DEBI ethics complaints and convenes (electronically or in person) on an ‘as needed’ basis or on request of ExCom. The panel also makes recommendations to ExCom with respect to ethics training programs for C-DEBI members. The Ethics Panel consists of Chair Karen Lloyd (Assistant Professor at U Tennessee), Frederick Colwell (Professor at Oregon State), Andrew Fisher (ExCom), Sharon Cooper (Education Officer of the IODP US Science Support Program at Lamont-Doherty Earth Observatory), and William Orsi (Assistant Professor at LMU Munich), representing several groups within C-DEBI. To date, the committee has not received any ethical complaints.

In addition, we continue to emphasize a comprehensive ethics policy for C-DEBI participants based on existing models starting with NSF and integrating with specific IODP and other institution policies. This sets forth a community standard to minimize and resolve conflicts effectively. The online ethics training is mandatory for all C-DEBI participants and completion by deadline is enforced.  See more at our [Ethics Policies webpage](http://www.darkenergybiosphere.org/about-our-center/ethics-policy/).

**External Evaluator**

The External Evaluator, Beth Rabin, assesses and evaluates the effectiveness of C-DEBI management, research, and education, outreach, and diversity programs and provides thorough, rigorous, independent, and results-based assessments to ExCom.

**Education & Outreach Steering Committee**

The Education & Outreach Steering Committee serves in an advisory role to the EOD Administration and also helps to review the small education and outreach grant proposals. The committee consists of current or previous STC Education and Diversity Directors Sharnnia Artis (UC Berkeley), Vanessa Green (Oregon Health & Science U), and Elisa Maldonado (UC San Diego).

**2. Management and Communications Systems**

C-DEBI is a distributed center, with members and participants around the world. Our Center and its participants have ample experience in long-distance collaboration and communication. There are weekly administrative and ExCom meetings via videoconferencing. We have a biweekly newsletter (sent to ~1000 e-mail addresses), a regularly updated website, and C-DEBI also has a presence on social media, including Facebook (cdebi) and Twitter (@deepbiosphere). C-DEBI’s annual meeting includes leadership and advisory groups, graduate and postdoctoral fellows, and invited guests. We also organize several targeted workshops annually and encourage members (especially postdoctoral scholars and early career scientists) to organize sessions at national and international meetings. Coordination of these communication activities is under the purview of the Administrative team.

The C-DEBI website plays a number of important roles in Center communications. As the website is often a first point of contact with the Center, numerous cosmetic improvements and performance enhancements have been implemented to improve user experience and first impressions, and this work will be ongoing as expectations for website usability continue to advance. The website also serves as the definitive source for Center goals, policies and programs, and here the documentation has been restructured to provide a clearer separation of concerns and to reflect changes to the Center’s foci as of Phase II. Going forward, we will continue to improve navigational awareness through greater visual differentiation of pages and entities, and promote the discoverability of related content via metadata enrichment. As a principal source of community news and activities, the website lists time-sensitive items on the front page and archives them for search. Working in conjunction with Diversity Director Noda, who has begun managing the Center’s social media presence, we will find ways for the Center’s multiple web outlets to leverage each other’s strengths in the coming year. Lastly, the website is intended to serve as a working resource for active research and education participants, and we have improved the website’s search interface and metadata-driven cross-linking. This year’s drive to centralize data assets in BCO-DMO has been a boon on this front – project data are now first level entities on the website – and future development will integrate BCO-DMO’s ontology-focused metadata system with our website interface and own metadata requirements. To lower technological barriers to data access and synthesis, we will leverage resources like iMicrobe, protocols.io, and other partners in the EarthCube community.

**4. Performance with Respect to the Strategic Implementation Plan**

Our leadership and management goal is to envision and enable the Center’s mission through inclusive and transparent decision-making; inspire Center members; and facilitate collaborative effort and guide all participants in the center via a cross-disciplinary and multi-institutional ethics program to instruct them on ethical and responsible conduct of scientific research. A fundamental challenge for C-DEBI leadership is to maintain trust and support among a diverse and interdisciplinary community of scientists, educators, and technologists. Maintaining confidence in STC leadership, throughout the existence of the Center, is essential if busy STC participants are to retain a willingness to allocate some of their limited time for advisory, review, and collaborative activities. STC leaders will also need to assure that there are robust opportunities for inter-institutional and cross-disciplinary exchanges and training, and help to secure external resources in support of ongoing and future STC activities. In addition, the Center will maintain a rigorous ethics training system for all C-DEBI participants and an Ethics Panel overseeing policies and procedures. Finally, STC leadership needs to remain focused on the critical goal that motivated formation of C-DEBI in the first place: creating a vibrant, innovative, and focused community, who will work together to achieve what cannot be accomplished by individuals working alone, to transform the nature of deep biosphere research.

**Target 1:** The decision-making process is defined, transparent and effective leading to a high degree of confidence, ownership, and engagement by STC participants in the Center.

|  |  |
| --- | --- |
| **Metric** | **Status/Problems** |
| Hold weekly administration meetings as well as weekly ExCom meetings and an annual ExCom face-to-face retreat to enable clear and effective management of the Center | Met |
| Survey the community every 1-2 years to establish effectiveness of leadership teams, decision making, and Center engagement with 70% of respondents rating leadership as being “clear/effective” or “very clear/effective” | Met (2015) |
| Invite the evaluation of Center research, education, diversity, and knowledge transfer management by the External Advisory Board (typically in conjunction with the C-DEBI annual meeting) for feedback and suggestions to the Director to improve the integration of C-DEBI programs and activities | Met |
| Update the C-DEBI Operations Manual to elucidate the functions of key individuals and groups and main research, education, outreach, and administration activities, programs, operations and procedures and post on the website with the Annual Report and Strategic Implementation Plan | Met |

**Target 2:** Communication is effective in facilitating the exchange of science, education of students, and promotion of other C-DEBI activities and opportunities.

|  |  |
| --- | --- |
| **Metric** | **Status/Problems** |
| Regularly update the comprehensive website at www.darkenergybiosphere.org with research and education portals and resources | Met |
| Distribute biweekly newsletters to C-DEBI community (participants and affiliates) to highlight recent and upcoming C-DEBI research and education programs and events and other relevant/partner activities and opportunities | Met |
| Continue to improve the private login site for internal documents and community reporting | Met |
| Solicit nominations for the next season of the videoconferenced Networked Speaker seminar series to present early career scientist research to the C-DEBI community | Met |
| Maintain protocol/procedure for issuance and usage of C-DEBI contributed publication numbers and of logo and branding information | Met |

**Target 3:** STC participants are engaged in cross-Center training and collaboration.

|  |  |
| --- | --- |
| **Metric** | **Status/Problems** |
| Organize 5-7 C-DEBI-specific opportunities for collaboration and training and entrain new membership (e.g., Center-wide Annual Meetings, Research Workshops, and Exchange Grants) | Met |
| Support 4-6 research and professional development opportunities specifically for graduate students and postdoctorals (e.g., workshops at Annual Meetings, professional development webinars, and fellowships and networking activities in Research and Education sections above) | Met |

**Target 4:** Community commitment to an environment promoting high ethical standards in the conduct of research is maintained.

|  |  |
| --- | --- |
| **Metric** | **Status/Problems** |
| Require 100% of participants complete ethics training within these standards | Met |
| Ethics Panel composed of Research, Education, ExCom and Early Career representatives resolves complaints regarding C-DEBI administration, funding and scientific conduct in a timely manner (within 6 months of being presented to C-DEBI) | Met |

**Target 5:** Strategies, tools, and resources are developed for sustainability of C-DEBI activities.

|  |  |
| --- | --- |
| **Metric** | **Status/Problems** |
| Secure $3M in aggregate (beyond initial STC funding) in support of C-DEBI activities | Met |

## 5. Plans for the Next Reporting Period

To further enhance C-DEBI’s culture of collaboration and cross-disciplinary thinking, we will continue to develop cyber-infrastructure for our website enabling public access and data sharing among the C-DEBI research community. The architecture for our online communities for collaboration and learning for has two principal objectives: 1) to support the connection among scientists and others in the C-DEBI project research community and 2) to foster the connections between C-DEBI scientists and educators. See also [Data Management and Integration Section IV](#_IV._DATA_MANAGEMENT_2).

# 

# VIII. CENTER-WIDE OUTPUTS AND ISSUES

## 1. Center Publications

In the current reporting period, the C-DEBI community produced 42 publications, including 39 peer-reviewed journal articles ([[Appendix J](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-J-Center-wide-Outputs.xlsx)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-I-Center-wide-Outputs.xlsx)). Details in the appendix include graduate and postdoctoral authors, contributing C-DEBI funding, expedition, site, habitat and theme association.

## 2. Conference Presentations and Other

##### Center participants reported 112 oral or poster presentations at venues including the 2016 Goldschmidt Conference in Tokyo, the 2015 and 2016 AGU Fall Meetings, and the 2016 ISME general meeting ([[Appendix J](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-J-Center-wide-Outputs.xlsx)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-I-Center-wide-Outputs.xlsx)).

## 3. Honors, Awards and Grants

##### C-DEBI participants reported receiving 34 (with another 4 pending) honors, awards and grants during the reporting year related to their C-DEBI funding ([[Appendix J](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-J-Center-wide-Outputs.xlsx)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-I-Center-wide-Outputs.xlsx)).

## 4. Placement of Graduated Students and Postdoctorals

##### Thirty-two C-DEBI undergraduate, graduate students, postdoctoral scholars, early-career scientists and educators obtained degrees or placement during the current reporting year ([[Appendix J](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-J-Center-wide-Outputs.xlsx)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-I-Center-wide-Outputs.xlsx)). C-DEBI funding contributing to the degrees or placement is identified.

## 5. Outputs of Knowledge Transfer Activities

The C-DEBI community developed 5 technologies in the current year including platforms and software. See also [Education Section III](#_III._EDUCATION_3) and [Data Management & Knowledge Transfer Section IV](#_IV._DATA_MANAGEMENT_3).

## 6. All Participants

Of the 286 individuals reported as being involved with Center activities, 140 are classified as “participants” (per NSF: individuals who have spent over 160 hours on Center activities), while 146 are “affiliates,” reported spending under 160 hours. Affiliates are included where they were reported as personnel on a C-DEBI grant or other budgeted item, attended a C-DEBI event, or have a titular role in the Center. Sources of Center support and known, subseafloor-related, event attendance are included per participant to provide further differentiation of engagement level. See [[Appendix J](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-J-Center-wide-Outputs.xlsx)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-I-Center-wide-Outputs.xlsx) for details.

## 7. Institutional Partners

C-DEBI has identified 92 participating institutions categorized per NSF reporting requirements. Types are determined based on the activities of its participants as follows:

Graduate student: *education, research*

Postdoctoral or researcher: *research*

Outreach or professional development: *education*

Program targets diverse groups: *diversity, education*

Participant worked on new tools, software, methods or products: *knowledge transfer*

Participant plays an advisory or managerial role in the Center: *all types*

Whether the institution has “participated” less or more than 160 hours is likewise determined by its affiliated participants. See [[Appendix J](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-J-Center-wide-Outputs.xlsx)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-I-Center-wide-Outputs.xlsx) for details. See also [External Partnerships Section V](#_V._EXTERNAL_PARTNERSHIPS_6).

## 8. Summary Table for Internal NSF Reporting Purposes

|  |  |
| --- | --- |
| Number of participating institutions (all academic institutions that participate in activities at the Center) | 87 |
| Number of institutional partners (total number of non-academic participants, including industry, states, and other federal agencies, at the Center) | 35 |
| Total leveraged support (funding for the Center from all sources other than NSF-STC) | $299,350 |
| Number of participants excluding affiliates (total number of people who utilize center facilities; not just persons directly supported by NSF) | 140 |

## 9. Media Publicity

##### Twenty-two media publicity items have been identified, including press releases, news articles, videos, and radio programming ([[Appendix J](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-J-Center-wide-Outputs.xlsx)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-I-Center-wide-Outputs.xlsx)).Additionally, C-DEBI has an active presence on Facebook (683 page likes) and Twitter (583 followers), and produces a bi-weekly newsletter (1,004 subscribers).

**10. Distributable Media**

##### Our [current brochure](http://www.darkenergybiosphere.org/wp-content/uploads/C-DEBIbrochure2016dec.pdf) provides an overview of the deep biosphere, C-DEBI research programs and themes, C-DEBI education and outreach programs, and how to get involved.

# IX. INDIRECT/OTHER IMPACTS

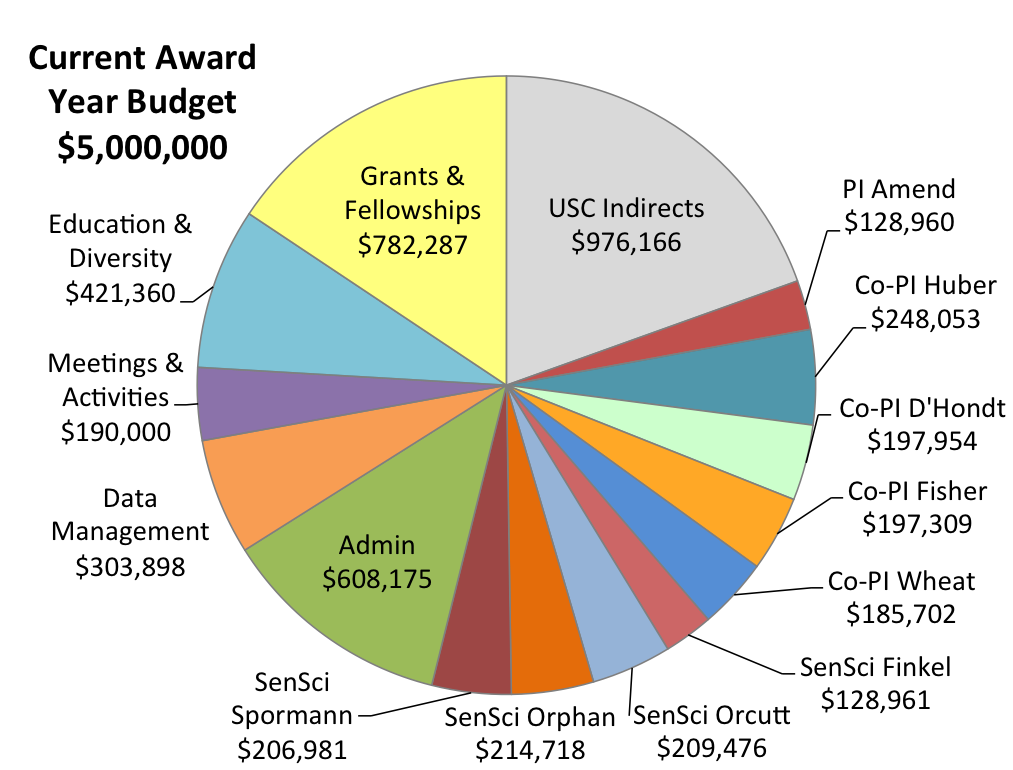
# 1. International Activities and Other Outputs, Impacts, or Influences

C-DEBI regularly and consistently engages the international community in the majority of its activities with individual and institutional partnerships as described throughout this report. Of particular note are research expeditions, especially IODP cruises, where multi-national participation is generally mandated, and C-DEBI-led conference sessions and workshops, where scientists from Germany, Japan, China, Great Britain, France, and other countries are commonly invited. All other outputs, impacts, or influences related to the Center’s progress and achievement in 2016 have been captured in other sections of this report.

# X. BUDGET

## 1. Current Award Year and Unobligated Funds

The Center’s current award year budget (4/1/16 – 3/31/17) is $5,000,000 with 34% supporting the research of the lead investigators including the PI, Co-PIs and Senior Scientists (see figure below). Indirect costs at USC consist of 20% of the budget. The remainder of almost half the budget serves the greater C-DEBI community with support for grants and fellowships, education and diversity programs, community meetings and activities, data management, and the general administrative operations based at USC. Our grants program includes support for small seed research grants up to $80,000-100,000 per year, and we are currently accepting proposals to begin in the next award year. This year we awarded 5 research grants of up to $80,000 each, 2 research exchange grants, 5 postdoctoral fellowships of ~$100,000 each and 2 graduate fellowships of ~$50,000 each (see details in the [Research Section II.2.i](#_i._Projects_from) and [[Appendix B](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-B-Active-Grants-and-Fellowships.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2015/12/Appendix-B-Active-Grants-and-Fellowships.pdf)). Our education and diversity programs target all audiences from teachers, K-12, undergraduates/community college students, graduates, postdoctorals, and the general public. Community meetings and activities include the C-DEBI Annual Meeting and C-DEBI Site Review. Administrative support includes salary and fringe benefits for the USC staff, work study students, travel and other operating costs. Education and diversity and data management staff are included in their respective categories.



As of October 31, 2016, we have expensed 60% of the total award of $5,000,000 for the current award year ([[Appendix K](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-K-Current-Award-Year-Budget.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-J-Current-Award-Year-Budget.pdf)). These expenditures of $2,978,381 consist of expenses posted in the USC ledger as of 10/31/16, however do not include liens/obligations (e.g., subcontracts) and known pending expenses (e.g., USC salaries). The bulk of the remaining funds will support operating costs (e.g., the upcoming Site Visit).

The discrepancy between what has been expensed (and reported to NSF by USC Sponsored Projects Accounting) and what is reported in [[Appendix K](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-K-Current-Award-Year-Budget.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-J-Current-Award-Year-Budget.pdf) is the inclusion of our committed expenses. For example, a large portion of our budget is in our small research grants and fellowships which are awarded as 1-2 year subcontracts or satellite accounts, and we report as committed expenses of ~$50K per award. However, the expenses reported by USC's SPA only include the individual invoices paid by USC which are incrementally billed/paid up to the entire award of ~$50K over the 1-2 year award period. What we report above carefully accounts for these types of commitments that may not be completely billed and paid for some period of time. See [[Appendix L](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-L-USC-Account-Status.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2015/12/Appendix-K-Management-Effectiveness-Evaluation.pdf) for a statement of our accounts in the USC Financial Accounting System.

## 2. Requested Award Year

In the next award period (4/1/17 – 3/31/18), we will continue to support the Co-PI and Senior Scientist research groups, central administration at USC, data management, meetings and activities, education and diversity programs and grants and fellowships (see figure below and [[Appendix M](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-M-Requested-Award-Year-Budget.pdf)](http://www.darkenergybiosphere.org/private/wp-content/uploads/2013/12/Appendix-L-Requested-Award-Year-Budget.pdf)).

# Macintosh HD:Users:Rosalynn:Dropbox:C-DEBI ADMIN:NSF REPORTING:Year 7_160401-170331:report:drafts:budget:requested year:requested pie.png

## 3. Center Support from All Sources

In addition to NSF core funds, the center received $299,350 directly from USC in the form of institutional returns on indirect costs this year, and will receive $280,423 in the requested award year (see table below). In the current award year and the requested award year, the USC Institutional Support funded additional Center activities including GEM Summer Course instructors (an additional 0.5 month of summer salary each), and additional C-DEBI community meetings and activities. Remaining funds are used at the Director’s discretion to support important Center projects and to position C-DEBI closer to renewal success. See details of non-monetary institutional commitment in [Appendix N](http://www.darkenergybiosphere.org/wp-content/uploads/Appendix-N-Institutional-Commitment.pdf).

|  |  |  |
| --- | --- | --- |
| **Center Support** | **Current Award Year** | **Requested Award Year** |
| NSF-STC Core Funds | $5,000,000 | $5,000,000 |
| USC Institutional Support | $299,350 | $280,423 |
| **TOTAL** | **$5,299,350** | **$5,280,423** |

**4. Additional PI Support From All Sources**

Additional levels of support not included above have been awarded to Center PIs from federal and state agencies, university, and private foundations and organizations (see table below).

|  |  |  |
| --- | --- | --- |
| **Additional Support** | **Current Award Year** | **Requested Award Year** |
| NSF | $760,464 | $1,155,714 |
| Other Federal Agencies  (NASA, NOAA, NIH) | $1,712,558 | $1,221,652 |
| State Government (California) | $179,163 | $303,500 |
| University | $80,180 | $80,180 |
| Private Foundations (DCO/Sloan, Simons) | $190,273 | $199,571 |
| **TOTAL** | **$2,922,638** | **$2,960,616** |