

U.S. DEPARTMENT OF COMMERCE

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PROJECT REPORT

VESSEL: NOAA Ship *Hi'ialakai*, Project HA-15-01, Leg 1

PROJECT

PERIOD: 21 January – 21 February, 2015

AREA OF

OPERATION: Phoenix Islands

TYPE OF

OPERATION: Personnel from the NOAA Pacific Islands Fisheries Science

Center's (PIFSC) Coral Reef Ecosystem Division (CRED), the NOAA PIFSC Scientific Operations, the NOAA Pacific Islands Regional Office, San Diego State University, and the U.S. Fish and Wildlife Service conducted interdisciplinary surveys of benthos, fishes, and physical oceanography in waters surrounding the Phoenix Islands as part of the National Coral Reef Monitoring Plan (NCRMP). All activities described in this report were covered by the following permits and authorizations: National Environmental Policy Act, Programmatic Environmental Assessment (PIFSC-20100901); Endangered Species Act, Section 7 consultation (PIR-2015-9580); U.S. Army Corps of Engineers, verification letter (POH-2009-00083); U.S. National Park Service, Scientific Research and Collecting Permit (NPSA-2015-SCI-0003); American Samoa National Marine Sanctuary, Research Permit (FBNMS-2014-003); U.S. Fish and Wildlife Service, Special Use Permit (12521-14001); American Samoa Department of Marine and Wildlife Resources, Scientific Study and Collection Permit (2014/010); Presidential Proclamations 8336 and 8337, respectively for Pacific Remote Islands and Rose Atoll Marine National Monuments



ITINERARY:

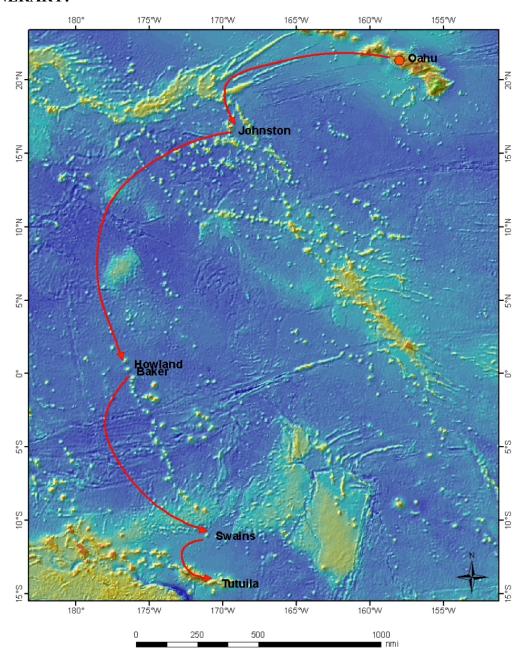


Figure 1. Track of NOAA Ship Hi'ialakai for the cruise HA-15-01, Leg 1, January 21-February 21, 2015, with Johnston Atoll, Howland Island, Baker Island, Swains Island, and Tutuila surveyed.

NOTE: This report only highlights work conducted around the Phoenix Islands during HA-15-01 Leg I. Due to their geographic and political distinction from the Phoenix Islands, all activities conducted around Swains and Tutuila Island during this leg are featured in a separate report.

Unless otherwise specified in the following daily summaries, these surveys occurred during each operational day: Rapid Ecological Assessment (REA) benthic surveys, REA fish surveys, towed-diver surveys; nearshore conductivity-temperature-depth (CTD) casts; water sample collections for dissolved inorganic carbon (DIC), total alkalinity (TA), and/or microbial analyses; and the deployments and recoveries of oceanographic instrumentation. Furthermore, shipboard multi-beam bathymetry data was collected daily at each island.

Jan 21	Fueling and overnight calibration of shipboard multibeam echosounders. Embark full science party.
Jan 22 – 24	Disembark: Frances Lichowski, Rhonda Suka, Jeremy Taylor (overnight multibeam calibration). Embark: Marie Ferguson, Louise Giuseffi, Kevin O'Brien. Transit to Johnston Atoll. Boat familiarization, station walkthroughs and dive neurological examinations for all scientists.
Jan 25	Arrive at Johnston Atoll. Transfer the U.S. Fish and Wildlife Service (USFWS) team via small boat to the island for terrestrial surveys.
Jan 26	Begin operations at Johnston Atoll. Deployed: 4 STRs. Retrieved: 4 STRs and 5 CAUs.
Jan 27	Continued operations at Johnston Atoll. Deployed and retrieved: 2 STRs and 5 CAUs.
Jan 28	Continued operations at Johnston Atoll. Deployed: 10 CAUs. Retrieved: 15 CAUs. Towed-diver surveys completed for Johnston Atoll.
Jan 29	Continued operations at Johnston Atoll. No oceanographic instruments deployed/retrieved. Towed-diver team shifted to conducting REA surveys for all remaining operational days here.
Jan 30	Recover USFWS team from the island. No other operations. Begin transit to Howland Island.
January 31 – Feb 2	Continue transit to Howland Island.
Feb 3	Arrive at Howland Island and begin operations. Transfer USFWS team via small boat to the island for terrestrial surveys. Only REA fish surveys were conducted today.
Feb 4	Continue operations at Howland Island. Deployed and retrieved: 3 STRs and 5 CAUs. Towed-diver surveys completed for Howland Island.

- Feb 5 Continue operations at Howland Island. Deployed: 3 STRs and 5 CAUs. Retrieved: 3 STRs and 8 CAUs. Towed-diver team shifted to conducting REA surveys for all remaining operational days here.
- Feb 6 Continue operations at Howland Island. Deployed: 5 CAUs. Retrieved: 4 CAUs.
- Feb 7 Continue operations at Howland Island. Deployed and retrieved: 5 CAUs. Begin overnight transit to Baker Island.
- Feb 8 Arrive at Baker Island and begin operations. Transfer USFWS team via small boat to the island for terrestrial surveys. Deployed: 3 STRs and 5 CAUs. Retrieved: 3 STRs. Towed-diver surveys completed for Baker Island.
- Feb 9 Continue operations at Baker Island. Deployed and retrieved: 3 STRs. Towed-diver team shifted to conducting REA surveys for all remaining operational days here.
- Feb 10 Continue operations at Baker Island. Deployed: 1 STR and 5 CAUs. Retrieved: 5 CAUs.
- Feb 11 Continue operations at Baker Island. Deployed and retrieved: 10 CAUs. Begin transit to Swains Island.
- Feb 12 21 Continue transit to Swains Island and Tutuila Island. Due to their geographic and political distinction from the Phoenix Islands, however, all activities conducted around Swains and Tutuila Island during this leg are featured in a separate report.
- Feb 21 Arrive at Pago Pago, Tutuila, American Samoa. End of HA-15-01 Leg I.

MISSION:

NOAA's Coral Reef Conservation Program (CRCP) has made the strategic decision to invest a portion of its annual operating budget in perpetuity to support a National Coral Reef Monitoring Plan (NCRMP), designed to assess and report the status and trends of environmental conditions, living reef resources, and the people and processes that interact with coral reef ecosystems. The NCRMP builds upon a decade of CRCP-supported, nationwide coral reef monitoring and reporting efforts, such as the Pacific Reef Assessment and Monitoring Program (Pacific RAMP), a CRED-led research program, and *The State of Coral Reef Ecosystems of the United States and Pacific Freely Associated States*, a NOAA Technical Memorandum compiled by the NOAA Center for Coastal Monitoring and Assessment. Although the scope of NCRMP is broad, it is intended to assess the status of coral reef ecosystems and their conditions throughout U.S.

states and territories and provide a steady and comprehensive analytical context to gauge changes in conditions at the sub-jurisdictional scale of an island or atoll.

The NCRMP focuses on four priority themes: climate change and ocean acidification; benthic communities (mainly corals); reef-associated fish communities; and socioeconomics. Biological monitoring for benthic and fish communities are conducted using a stratified random sampling design throughout shallow-water (0–30 m), hard-bottom coral reef habitats. Monitoring of climate change and ocean acidification is achieved by means of sustained, remotely sensed and *in situ* observations of ocean temperature; autonomous and discrete water sampling for analyses of near reef and surface seawater carbonate chemistry; and distinct biological installations designed to provide integrated, ecosystem-wide response data (e.g., biodiversity, calcification, and bioerosion) in the context of climate change. In the Pacific, biological (benthic and fish) and climate monitoring are conducted on a triennial basis. Socioeconomic monitoring is led by the CRCP at headquarters in Silver Spring, MD, and stands outside the scope of the NCRMP monitoring and assessment expeditions; therefore, it is not addressed in this cruise report.

The goals of the HA-15-01 Pacific Reef Assessment and Monitoring Cruise are as follows:

- A. Conduct ecosystem monitoring for benthic cover (community structure), coral populations (species composition, abundance, size distribution, and condition), and fish populations (species composition, abundance, and size distribution) of the shallow-water (≤ 30 m) coral reef ecosystems of American Samoa as well as the U.S. Pacific Remote Island Areas (PRIAs) of the Phoenix and Line Islands.
- B. Deploy and retrieve a suite of instruments and installations—including subsurface temperature recorders (STRs), CTDs, acoustic Doppler current profilers (ADCP), pH sensors (SeaFET), programmable underwater collectors (PUC), autonomous reef monitoring structures (ARMS), calcification accretion units (CAUs), and bioerosion monitoring units (BMUs) to allow for remote, long-term monitoring of oceanographic, environmental, and ecological conditions of the coral reef ecosystems of American Samoa as well as the U.S. Pacific Remote Island Areas (PRIAs) of the Phoenix and Line Islands.
- C. Conduct shallow-water CTD hydrocasts and collect discrete water samples for DIC, TA, and microbial community analyses to depths ≤ 30 m to examine the chemical, physical and biological linkages supporting and maintaining these island ecosystems.
- D. Conduct shipboard multibeam observations to examine the seafloor contours in areas that were previously poorly mapped and collect continuous ADCP, sea surface temperature (SST), salinity, and fundamental meteorological data (air temperature, wind speed and direction, barometric pressure, and relative humidity).

E. Collect a small number of shallow-water and coral rubble samples to examine microbe abundance, diversity, and function.

RESULTS:

This section provides operational totals regarding research activities (Table 1), specifics regarding data collected during cruise HA-15-01 Leg I, and a summary of important observations made while at sea. Note that results for Legs II, III, and IV for this cruise are detailed in separate reports. For more information pertaining to the data collected, methodology employed at the islands visited, see Appendices A–E.

Research Activity	JOH	HOW	BAK
Scuba Dives	176	200	203
Towed-diver Surveys: Benthic and Fish	16	5	5
Combined Length (km) of Towed-diver Surveys	31.5	11.2	11.5
REA Sites: Benthic	15	21	15
REA Sites: Fish	31	35	36
Microbial Samples	6	7	7
Microbial Metagenome Samples	3	2	2
CAUs Retrieved	25	22	15
CAUs Deployed	15	20	20
STRs Retrieved	6	6	7
STRs Deployed	6	6	7
Shallow-water CTD Casts	10	8	10
Shallow-water DIC Water Samples	11	9	10

The coral reef ecosystems of American Samoa and the U.S. Pacific Remote Island Areas (PRIAs) of the Phoenix and Line Islands have been surveyed biennially since 2002 and triennially starting in 2012 through CRED's Pacific RAMP. The cruise HA-15-01 marked this program's seventh expedition around the islands and atolls of Johnston Atoll, Howland Island, Baker Island, Swains Island, Tutuila, Ofu-Olosega, Tau, Rose Atoll, Jarvis Island, Palmyra Atoll, and Kingman Reef. Herein, we present highlights from our observations of Leg I of the expedition (Johnston Atoll, Howland Island, and Baker Island).

Johnston Atoll:

- Teams reported frequent sightings of overturned Acropora tables throughout the atoll. Furthermore, tow team reported that much of the coral on the Northwest side of the atoll experienced recent damage (e.g., fragmented or dislodged), likely as a result of a recent large North swell.
- USFWS observed a grounded private sailboat, which our ocean and climate change team towed out to safety.

- Benthic team observed two coral species listed as threatened under the Endangered Species Act (ESA): *Acropora speciosa* and *Acropora retusa*
- A large (200 cm) *Carcharhinus galapagensis* shark was seen on the northwest side of the island.
- Several moderate- to large-sized (130 190 cm) *Carcharhinus amblyrhynchos* sharks were seen northwest and southwest sides of the island.
- Multiple large schools (up to 300 individuals) of the brown surgeonfish *Acanthurus nigroris* were observed around the atoll.

Howland Island:

- A total of 7 manta rays were present on one fish REA dive. USFWS even reported seeing manta rays from land "surfing the waves". This level of abundance is anecdotally consistent with what has been reported here before.
- Tow team noted an abundance of the corralimorph, Rhodactis sp., on the Northwest and West side of the island. This matting benthic cnidarian has been spotted in these areas on recent ASRAMP cruises and appears to be holding steady.
- Benthic team saw a blue marlin on their safety stop and also spotted *Acropora retusa*, one of the coral species on the ESA threatened species list.
- In addition, the benthic team conducted one of their REA surveys at a large mono-stand of *Porites rus*, with one colony measuring over 2.5 meters, perfectly intact with no partial mortality.

Baker Island:

- Similar to previous survey years, the benthic team observed large thickets of branching *Acropora* sp. on the east side of the island.
- Fish team observed large schools of holocentrids and acanthurids, as well as the humphead wrasse, *Cheilinus undulatus*, on the southeast side of the island.

The following monitoring data and samples were collected during this expedition:

Ocean and Climate Change Monitoring Oceanographic Instrumentation and Biological Installations:

- STR recoveries/deployments at 1, 5, 15, 25 m depths
- Recoveries/installations of ARMS for assessment of cryptic invertebrate biodiversity present within the coral reef
- Recoveries/installations of CAUs to for assessment of CaCO₃ deposition rates
- Recoveries/installations of BMUs for assessment of bioerosion rates

Nearshore Oceanography from Small Boats:

- Shallow-water CTD hydrocasts to depths ≤ 30 m, including all sites where CAUs and discrete water samples were collected for DIC and TA
- Collection of water samples for DIC and TA analyses (collected in concert with shallow-water (≤ 30 m) CTD hydrocasts) and further calculations of the carbonate chemistry found within the water column and at the reef

Shipboard Oceanography:

- Shipboard ADCP transects for real-time measurements of ocean current direction and magnitude
- Real-time collection of meteorological data: solar radiation, air temperature, barometric pressure, and wind speed and direction
- Real-time measurments of seasurface temperature and salinity using a flow-through system onboard the ship

Biological Monitoring

REA Benthic Surveys:

- Digital still photographs of overall site character and typical benthos
- Digital still photographs of the benthos along transect lines
- Number, species or genus, size, and condition of all coral colonies observed within belt transects of known area
- Digital still photographs of diseased corals and coralline algae
- Water samples and benthic rubble grabs at select REA sites for microbial studies

REA Fish Surveys:

- Number, species, and estimated sizes of all fishes observed within visually estimated 7.5 m radius, stationary-point-count surveys
- Visual estimates of benthic cover, habitat type, habitat complexity, and urchin density
- Digital still photographs of the benthos along transect lines

SCIENTIFIC PERSONNEL:

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APPENDIX A: METHODS

This appendix describes the methods and procedures used by the Coral Reef Ecosystem Division (CRED) of the NOAA Pacific Islands Fisheries Science Center during its Pacific Reef Assessment and Monitoring Program (Pacific RAMP) cruise HA-15-01 on the NOAA Ship *Hi`ialakai* during the period of January 21 – May 3, 2015.

A.1. OCEAN AND CLIMATE CHANGE

(Jeanette Clark, Kevin O'Brien, Noah Pomeroy)

Three main activities were conducted for the monitoring of climate and ocean change: (1) near-shore oceanographic and water quality surveys; (2) deployment and retrieval of an array of subsurface moored instrumentation and installations to provide continuous, high-resolution time-series of physical observations or integrated, ecosystem-wide biological process data; (3) shipboard meteorological observations, including wind speed and direction, relative humidity, air temperature, and barometric pressure.

Climate and ocean acidification monitoring efforts at each survey site fall into four complementary levels of increasing resolution. These are intended to document the island-scale, water chemistry, spatial and temporal variability of reef water thermal structure across a depth gradient, and the integrated biological responses of the reef community to the prevailing chemical and physical conditions.

- 1- Class 0 sites: Only discrete water samples are collected and analyzed for dissolved inorganic carbon (DIC) and total alkalinity (TA).
- 2- Class 1 sites: Only subsurface temperature recorders (STRs), SBE 56 temperature loggers (Sea-Bird Electronics, Inc., Bellevue, Wash.), are deployed.
- 3- Class 2 sites: Include collection of discrete water for DIC and TA; STR deployments; benthic community surveys and benthic still photograph records. Biological installations, including Calcification Accretion Units (CAUs), Bioerosion Monitoring Units (BMUs), Autonomous Reef Monitoring Structures (ARMS), and coral coring are added.
- 4- Class 3 sites: A MAPpCO2 buoy system is added to the Class 2 site setup.

Most of the CRED's efforts focus on establishing Class 0 and Class 2 sites at select locations distributed along the four cardinal directions around each island surveyed. For "Class 2 sites" and above, thermal structure measurements are obtained based on the deployment of subsurface temperature recorders (STRs; SBE 56) along a perpendicular forereef transect at 1, 5, 15, and 25 m depth; each SBE 56 records the near-reef water temperature at the same time, on a 5 minute interval, for the duration of the instrument's deployment. Within this context, a permanent water quality, temperature, and biological survey/sampling site, designated as a *NCRMP Monitoring Station*, is established at the

15m depth STR location, at select islands. In addition to the SBE 56 the NCRMP Monitoring Station includes: deployment of 3 ARMS units, 5 CAUs, and 5 BMUs; collection of 3 carbonate chemistry water samples (with associated CTD casts); acquisition of still photographic benthic imagery to document benthic cover and composition; and rugosity measurements of benthic topographic complexity.

A.1.1. Moored Instruments for Time-series Observations

CRED accomplishes long-term oceanographic assessment and monitoring through the deployment and retrieval of a variety of platforms, which either electronically record *insitu* measurements (temperature, currents, and waves) or by facilitating biological recruitment/growth on fabricated structures. The following types of oceanographic instruments and biological installations were retrieved or deployed during this cruise.

Subsurface Temperature Recorder (STR): STRs are deployed at select sites to provide high-resolution (5-min interval) temperature data.

Ecological Acoustics Recorder (EAR): EARs are long-term recorders that monitor the sounds of marine animals and vessel traffic around the islands. Previously deployed EARs will be retrieved

Calcification Accretion Unit (CAU): CAUs are used to detect changes in calcification rates and net accretion of crustose coralline algae and other benthic sessile calcifiers. Five CAUs are deployed at each selected location.

Bioerosion Monitoring Unit (BMU): BMUs provide a proxy for an integrated signal of net reef bioerosion. Five CAUs are deployed at each selected location.

Autonomous Reef Monitoring Structure (ARMS): ARMS provide an assessment of cryptic taxonomic diversity of coral reef associated species. Three ARMS are deployed at each selected location.

A.1.2. Hydrographic Surveys

Detailed oceanographic and water quality surveys were conducted using the following sampling techniques and equipment.

Shallow-water (Near-shore) Conductivity, Temperature, and Depth Casts: a CTD profiler deployed from a small boat provided water column data on temperature, conductivity, which is related to salinity, and pressure, which is related to depth (SBE 19plus SeaCAT Profiler). A transmissometer (C-Star, WET Labs, Philomath, Ore.) provided profiles of beam transmittance, which is related to turbidity. A dissolved oxygen sensor (SBE 43, accuracy of 2% of saturation) also was attached and measurements were made in concert with CTD measurements. A CTD cast was performed at each location where a water sample was collected. The CTD is lowered by hand, off a small boat at descent rates of ~ 0.5–0.75 m s-1 to depths up to 30 m.

Shipboard Acoustic Doppler Current Profiler (ADCP): a ship-based sensor provided transects of directional ocean current data (75-kHz Ocean Surveyor, Teledyne RD Instruments Inc., Poway, Calif.). The system was configured with an 8-m pulse length, 16-m depth bins starting at 25 m and extending typically to 600 m (range depended on density and abundance of scatterers), and 15 min averaged ensembles.

Water Chemistry: water samples for analyses of concentrations of dissolved inorganic carbon (DIC) and Total Alkalinity (TA) were collected at select locales concurrently with CTD casts.

A.2. BENTHIC AND MICROBIAL COMMUNITIES

(Hatsue Bailey, Marie Ferguson, Joao Garriques, Brett Schumacher, Cynthia Silveira, Aviv Suan, Dione Swanson, Jesse Tootell)

A two-stage stratified random sampling design was employed to survey the Rapid Ecological Assessment (REA) sites in the Phoenix Islands and Swains Island. The survey domain encompassed 99% of the mapped area of reef and hard bottom habitat., . Both reef zone (lagoon, back reef , and fore reef) and depth categories of shallow (0-6 m), moderate (>6-18 m) and deep (>18-30 m) were incorporated into the stratification scheme. Allocation of sampling effort was proportional to strata area. Sites were randomly selected within each stratum.

A.2.1. Benthic Composition and Coral Demographics

Surveys at each site were conducted within two, 10 m^2 belt transects. Adult coral colonies ($\geq 5 \text{ cm}$) were surveyed within four ($1.0 \times 2.5 \text{ m}$) segments in the following manner: 0–2.5 m (segment 1); 5.0–7.5 m (segment 3); 10–12.5 m (segment 5); and 15–17.5 m (segment 7). All colonies whose center fell within 0.5 m on either side of each transect line were identified to lowest taxonomic level possible (species or genus), measured for size (maximum diameter to nearest cm), and morphology was noted. In addition, partial mortality and condition of each colony was assessed. Partial mortality was estimated as percent of the colony in terms of old dead and recent dead and the cause of recent mortality was identified if possible. The condition of each colony including disease and bleaching was noted along with the extent (percent of colony affected) and level of severity (range from moderate to acute).

Juvenile coral colonies (< 5 cm) were surveyed within 10 m^2 along the same two transects as adults, specifically within three ($1.0 \times 1.0 \text{ m}$) segments: 0-1.0 m (segment 1); 5.0-6.0 m (segment 3); and 10.0-11.0 m (segment 5). Juvenile colonies were distinguished in the field by a distinct tissue and skeletal boundary (not a fragment of larger colony). Each juvenile colony was identified to lowest taxonomic level (genus or species) and measured for size by recording both the maximum and perpendicular diameter to the nearest 2 mm.

Still photographs were collected to record the benthic community composition at predetermined points along the same 2 transect lines with a high-resolution digital camera mounted on a pole. Photographs were taken every 1 m from the 1 m to the 15 m mark. This work generates 30 photographs per site, which are later analyzed by CRED staff and partners using the computer program Coral Point Count with Excel extensions (CPCe). This analysis is the basis for estimating benthic cover and composition at each site (benthic habitat photographs at sites surveyed by the fish team are also analyzed).

In addition to site-specific REA surveys, broad-scale towed-diver surveys were used to determine the benthic composition of shallow-water habitats around each island and to quantify the abundance of target macroinvertebrates, including crown-of-thorns seastars (COTS), sea urchins, sea cucumbers, and giant clams. A pair of divers, by means similar to a manta-tow technique, were towed 60 m behind a small boat, a 6-m survey launch from SAFE Boats International (Port Orchard, Wash.), with one diver quantifying the benthos and the other quantifying fish populations. Each towed-diver survey lasted 50 min, broken into ten 5-min segments, and covered ~ 2 km. To georeference the survey launch's track, latitude and longitude coordinates were recorded at 5-s intervals using a Garmin GPSMap 76 global positioning system (GPS) unit on the boat. A custom algorithm was used to calculate the track of the divers based on speed and course of the boat and depth of the diver. Each towed-diver platform, or towboard, was equipped with an SBE 39 temperature and depth sensor programmed to record at 5-s intervals. At the end of each day, data were downloaded, processed, and presented in ArcGIS and can be displayed in conjunction with IKONOS satellite imagery, NOAA chart data, or other spatial data layers.

Towed-diver benthic surveys recorded habitat type and complexity; percentages of cover of benthic fauna, including hard corals, stressed hard corals, octocorals, macroalgae, and crustose coralline red algae, and of physical features, including sand and rubble; and counts of target macroinvertebrates and marine debris. Towed divers classified percentage of cover using a system of 10 bins, ranging from 0% to 100% cover of the benthos. Target macroinvertebrates were counted up to 25 individuals per segment and then binned into larger groups when exceeding 25. The benthic towboard was equipped with a downward-facing, high-resolution digital still camera. The camera took a photograph of the substrate every 15 s. These photos, like the SBE 39 data, are linked spatially with GPS track files taken aboard the survey launch.

A.2.2. Microbial Communities

As part of the ongoing effort to understand the microbial community, two types of water samples were collected. The first type included two diver-deployable Niskin bottles that were used to collect water at REA sites of moderate depth. The Niskin bottles (two 2-L replicates) were filled with "reef water" collected from < 1 m above the benthos. These water samples were returned to the ship and processed for DOC, particulate organic matter (POM), nutrients, microbial (Bacteria and Archaea) and viral counts (fluorescent microscopy), fluorescence-activated cell sorting (FACS, heterotrophs vs autotrophs), and microbial and viral community composition (coarse analysis: 16s rRNA).

The other type of water collection was for metagenomic analysis of the microbial and viral community associated with reef benthos. Only one sample per island was procured. This collection involved carboys (four 20L replicates) that were also filled with "reef water" collected from < 1 m above the benthos. The samples were collected using a flexible, plastic hose with a carboy bottle attachment. The carboys were filled using a small, lightweight pump attached to the other end of the hose.

The following data items were collected (for reef- and pore-water samples):

- DOC: 2 replicates
- POM: 2 replicates
- Nutrients: 2 replicates
- Microbial (Bacteria and Archaea) and viral abundance: 2 replicates (0.02-μm filters, stained using SYBR Gold, Molecular Probes Inc., Eugene, Ore.)
- Microbial (Bacteria and Archaea) size structure : 2 replicates (0.2-μm filters, stained using 4', 6-Diamidino-2-phenylindole (DAPI))
- Microbial community composition (FACS, heterotrophs/autotrophs): 6 replicates
- Microbial community composition (16s rRNA): 1 (0.22-µm filters)
- Microbial community composition (metagenome): 1 sample, (3–6 filters of 0.45 μm)
- Viral community composition (metagenome): 1 sample, (3–6 vials)
- Coral rubble or sediment: 6 replicate bags

Processing of Water Samples: This section describes the techniques used to process the water samples.

Enumeration of microbes and viruses. Samples of 1-mL from each Niskin were fixed using paraformaldehyde and stained using the general nucleic acid stain SYBR Gold. The samples were filtered through 0.02-μm Anodisc filters and mounted on a microscope slide. Direct counts of microbes and VLPs will be completed using fluorescent microscopy and Image Pro software.

Microbial biomass. Samples of 1-mL from each Niskin were fixed using glutaraldehyde and filtered through 0.2- μm filters. These filters were stained with DAPI, a general nucleic acid stain for staining double-stranded DNA (dsDNA) that allows length and width data to be obtained for individual microbes. These filters were then mounted on a microscope slide for analysis using fluorescent microscopy and Image Pro software. Slide analysis will be performed at San Diego State University (SDSU). All filters were stored at -20° C for archival purposes.

Enumeration of autotrophic vs. heterotrophic microbes: Flow cytometry will be used to assess the ratio of autotrophic to heterotrophic microbes in the water column. This technique also will provide complementary data for microbial abundance, microbial community structure, and levels of chlorophyll-a.

Six 1-mL samples of water from each REA site were pushed through a 20- μ m filter. This filtrate was dispensed into cryovials (6 × 1 mL) and fixed with glutaraldehyde. Vials were inverted to mix. Glutaraldehyde-preserved samples were flash frozen in liquid nitrogen contained in a dry shipper to prevent damage to microbial cells. These samples will be shipped on dry ice to SDSU for flow cytometry analysis.

Water Chemistry (DOC/POC): 30 mL of seawater were filtered through a 47 mm precombusted glass fiber filters from each of the 2 Niskin bottles, and the filtrate was collected in pre-combusted plastic bottles. The bottles were stored at −20°C. To assess POC, 500 mL of seawater were filtered through a 47 mm pre-combusted glass fiber filter (2 replicates), and the filters were stored at −20°C. Stable isotopes of carbon and nitrogen also will be analyzed from the filters via standard protocols at SDSU.

Collection of DNA for metagenomics: The community structure of the microbes and viruses associated with the water column was assessed by metagenomic analysis. Metagenomics is a powerful tool for studying environmental populations as < 1% of all environmental microbial diversity is currently cultivable. The steps for analysis of microbial community diversity and function involve collection of environmental DNA followed by 16S rRNA gene sequencing. Approximately 1.5–2 L of reef water was filtered through a 0.22 μm sterivex filter. DNA isolation and metagenomic analysis will be completed at SDSU.

At one REA site per island, four 20-L collapsible carboys of water were filled with water from reef crevices or reef benthos using a manual bilge pump. Upon return to the ship, this water sample was pre-filtered through 100-µm mesh and concentrated using tangential flow filtration (TFF). TFF concentrates the bacteria and viruses in the water, bringing the initial 70–80 L of water to a final volume of ~ 500 mL. This concentrate was then filtered through 0.45-µm filters to capture microbes (Bacteria and Archaea). These filters were frozen at –20°C. The DNA of the entire community will be extracted and sequenced at SDSU, and the diversity and function of the microbial communities associated with the reef benthos will be analyzed. The filtrate from this sample contains concentrated viruses. Chloroform was added to this filtrate to kill any small microbes that passed through the 0.45-µm filter, and the sample was stored at 4°C. Once shipped to SDSU, viruses will be isolated from the viral concentrate, and community DNA will be extracted and sequenced. This extracted and sequenced DNA will then be analyzed for viral community diversity and function.

Collection of Benthic Samples (if time permits): This section describes samples, or benthic grabs, collected if time permitted.

Collection of benthic microbial DNA: In addition to changes in the microbial community associated with the water column, we are also interested in whether or not community shifts in microbes associated with the benthos are a useful indicator of reef health. When time permits, six fistfuls of coral rubble or sediment and six pieces of the most dominant algal-type will be collected in Ziploc bags. Both the algal and rubble/sediment samples

were frozen at -20°C. These samples stayed on the ship until it returned to Honolulu. The bacterial 16s rRNA genes associated with these samples will be sequenced to characterize the microbial communities associated with the benthos (rubble and algae).

A.3. REEF FISH COMMUNITIES

(Jacob Asher, Paula Ayotte, Louise Giuseffi, Andrew Gray, Kelvin Gorospe, James Morioka)

Divers conducted REA fish surveys using the stationary-point-count (SPC) method at pre-selected REA sites. Two separate teams performed these surveys. Each team consisted of 2 divers, and conducted 1 SPC survey per site. All fish REA sites visited were selected using a stratified random sampling design in shallow (0–6 m), moderate (6–18 m), or deep (18–30 m) depth strata, in the forereef habitat strata. Surveys were performed using a 30-m transect line set along a single depth contour. The REA sites selected for fish surveys typically differ in location from the REA sites where benthic surveys were conducted.

Once a transect line was deployed, the 2 divers moved to the 7.5-m and 22.5-m marks on this transect line to start their SPC surveys. Each of these marks or points, with 1 diver at each, served as the center of a visually estimated cylindrical survey area with a radius of 7.5 m. During the first 5 min, divers only recorded the presence of species within their respective cylinders. Afterwards, divers went down their respective species lists, which were created from their work during the initial 5 min of a survey, sizing and counting all individuals within their cylinder, one species at a time. Cryptic species missed during the initial 5 min of a survey could still be counted, sized, and added to the original species list. Fish species observed at a REA site but not recorded during the SPCs were recorded for presence data.

After a survey was completed, divers recorded benthic habitat information within their respective cylindrical survey areas. Divers visually estimated habitat complexity, habitat type, and percentage of cover for hard corals, macroalgae, crustose coralline red algae, turf algae, and sand. Urchin densities were also estimated. Every meter along the transect line, still photographs were taken of the benthos to the right side of the line. This work generates 30 photographs per site, which together with the habitat photographs at sites surveyed by the benthic team, are later analyzed, implementing Coral Point Count with Excel extensions (CPCe), to estimate the benthic cover and composition at each site.

In addition to site-specific REA surveys, broad-scale towed-diver surveys (described above under section: A.2.1. Benthic Composition and Coral Demographics) were used to characterize the communities of large-bodied, highly-mobile fish of the shallow-water habitats around each island.

Towed-diver fish surveys record, to the lowest possible taxon, all fishes > 50 cm in total length along a 10-m swath during each 5-min segment. Individual fishes were counted and their species (or lowest possible taxon) and length in centimeters recorded. Sightings of species of particular concern observed outside the survey swath were classified as

presence/absence data and were recorded separately from the quantitative swath data. At the end of each day, data were transcribed from field data sheets into a centralized Microsoft Access database. Biomass values are calculated using species-specific length-weight parameters and are normalized by area (i.e., kg 100 m⁻²). The fish towboard was equipped with a forward-looking digital video camera that created a visual archive of the survey track that can be used to evaluate stochastic changes in reef environments, particularly following episodic events, such as coral bleaching and grounding of a vessel.

APPENDIX B: JOHNSTON ATOLL

Johnston Atoll, located at 16°45′ N, 169°31′ W in the central Pacific, includes four islands and a small lagoon and is part of the Pacific Remote Islands Marine National Monument. For information about the methods used to perform the activities discussed in this appendix, please see Appendix A: "Methods." In addition to the activities described in this appendix, a U.S. Fish and Wildlife Service field party went ashore to Johnston Atoll during HA-15-01, Leg I, to conduct surveys of terrestrial flora and fauna.

B.1. Ocean and Climate Change

Oceanographic operations during the cruise HA-15-01 at Johnston Atoll entailed numerous retrievals and deployments of oceanographic moored instruments including the installation of subsurface temperature recorders (STRs) and calcification acidification units (CAUs), as well as nearshore water sampling and conductivity, temperature, and depth (CTD) casts and acoustic Doppler current profiler (ADCP) transects.

Seven shallow-water CTD casts were performed at locations where water samples were collected. Eight shallow-water samples were collected for analysis of dissolved inorganic carbon (DIC), total alkalinity (TA), and salinity. In addition, 6 STRs and 15 CAUs were retrieved, and 6 STRs and 25 CAUs were deployed. (Fig. B.1.1 and Table B.1.1).

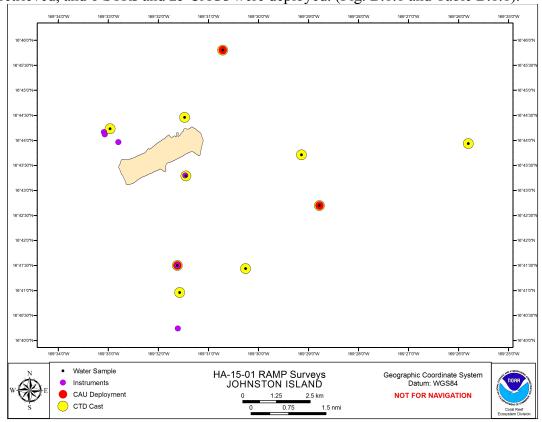


Figure B.1.1.--Mooring sites where oceanographic instruments and biological installations were retrieved or deployed and locations of nearshore CTD casts and water sampling performed at Johnston Atoll during cruise HA-15-01.

Table B.1.1.- Geographic coordinates and depths of the moored oceanographic instruments (STRs) and biological installations (CAUs), that were retrieved or deployed at Johnston Atoll during cruise HA-15-01.

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		Instrument			Depth		
Site	Date	Туре	Latitude	Longitude	(m)	Retrieved	Deployed
JOH_001	26-Jan-15	STR	16.73280	-169.54670	1.2	1	1
JOH_013	26-Jan-15	STR	16.72170	-169.52435	5.2	1	1
JOH_014	26-Jan-15	STR	16.73615	-169.55146	24.7	1	1
JOH_015	26-Jan-15	STR	16.73536	-169.55117	14.9	1	1
JOH_012	27-Jan-15	STR	16.67068	-169.52682	25.0	1	1
JOH_016	27-Jan-15	STR	16.69167	-169.52700	15.2	1	1
JOH-11	26-Jan-15	CAU	16.72150	-169.52412	11.9	5	0
JOH-12	27-Jan-15	CAU	16.74755	-169.52406	11.6	5	0
JOH-56	27-Jan-15	CAU	16.69167	-169.52699	15.2	0	5
JOH-07	28-Jan-15	CAU	16.71160	-169.47961	12.2	5	5
JOH-09	28-Jan-15	CAU	16.72856	-169.48563	7.6	5	0
JOH-10	28-Jan-15	CAU	16.76343	-169.51190	14.6	5	5

B.2. Benthic and Microbial Communities

REA benthic survey sites were chosen using a stratified random design. Belt-transect surveys were conducted at 16 REA sites at Johnston Atoll. Water samples for microbial analyses were collected at 6 sites (Fig B.2.1; Table B.2.1). For more information about collections, see Table E.1.1 in Appendix E: "Biological Collections."

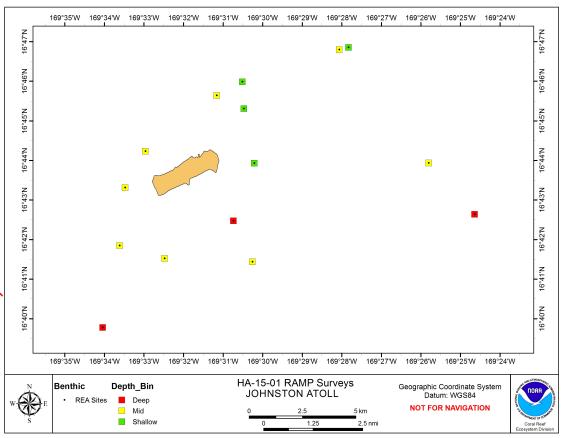


Figure B.2.1.-Locations of REA benthic sites surveyed at Johnston Atoll during cruise HA-15-01. All of these REA sites were selected using a stratified random design.

Table B.2.1.—Summary of the REA benthic surveys and microbial water collections performed at Johnston Atoll during cruise HA-15-01.

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		Depth	Reef	Depth			REA Coral	Microbial
REA Site	Date	Bin	Zone	(ft)	Latitude	Longitude	Survey	Samples
JOH-11	26-Jan-15	Mid	Lagoon	39	16.7215	-169.52412		Х
JOH-701	26-Jan-15	Mid	Forereef	40	16.72189	-169.558	x	
JOH-776	26-Jan-15	Mid	Forereef	49	16.73723	-169.5494	х	
JOH-711	26-Jan-15	Deep	Forereef	63	16.70791	-169.51231	х	
JOH-664	27-Jan-15	Deep	Forereef	78	16.66298	-169.56743	х	
JOH-1999	27-Jan-15	Deep		82	16.67068	-169.52682		Х
JOH-56	27-Jan-15	Mid		50	16.69167	-169.52699		Х
JOH-715	27-Jan-15	Mid	Forereef	61	16.69206	-169.54137	х	

JOH-722	27-Jan-15	Mid	Forereef	50	16.69064	-169.50428	x	
JOH-765	27-Jan-15	Mid	Forereef	40	16.69749	-169.56025	x	
JOH-675	28-Jan-15	Deep	Forereef	86	16.71064	-169.4107	х	
JOH-09	28-Jan-15	Mid	Lagoon	30	16.72856	-169.5119	х	Х
JOH-714	28-Jan-15	Mid	Forereef	48	16.73229	-169.43002	х	
JOH-833	28-Jan-15	Shallow	Forereef	13	16.75514	-169.50791	х	
JOH-843	28-Jan-15	Shallow	Lagoon	21	16.73222	-169.50352	х	
JOH-10	28-Jan-15	Mid	Lagoon	48	16.76343	-169.5119		Х
JOH-610	29-Jan-15	Shallow	Backreef	22	16.78093	-169.46381	х	
JOH-12	29-Jan-15	Mid	Lagoon	36	16.74103	-169.52457		Х
JOH-643	29-Jan-15	Mid	Backreef	35	16.78001	-169.46771	х	
JOH-645	29-Jan-15	Shallow	Backreef	21	16.76645	-169.50857	Х	
JOH-613	29-Jan-15	Mid	Backreef	35	16.76069	-169.5194	Х	

Additionally, during the HA-15-01 cruise, 16 towed-diver surveys were completed around Johnston Atoll, covering a total length of 31.5 km of the ocean floor (Fig. B.2.2).

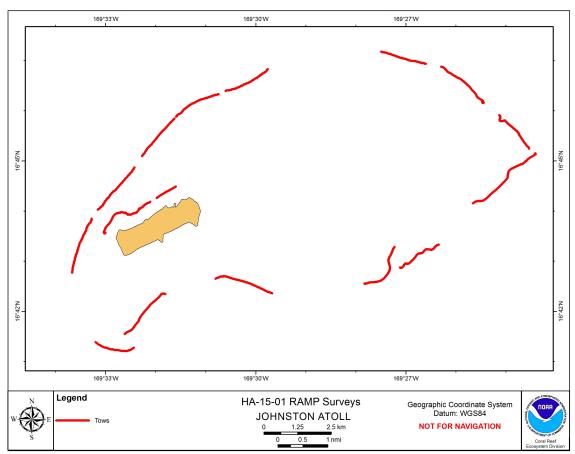


Figure B.2.2.-Track locations of towed-diver surveys conducted at Johnston Atoll during the cruise HA-15-01.

B.3. Reef Fish Communities

REA fish survey sites were chosen using a stratified random design. Stationary-point-count surveys were conducted at 31 REA sites at Johnston Atoll over five different habitat strata: deep, and moderate forereef, and deep, moderate, and shallow lagoon (Fig. B.3.1 and Table B.3.1). No fishes were collected during these surveys.

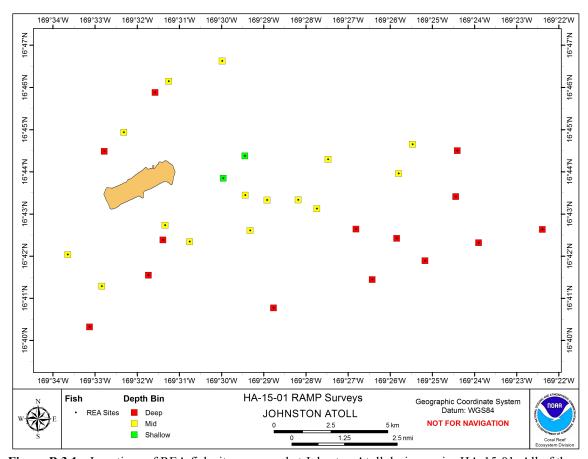


Figure B.3.1.-Locations of REA fish sites surveyed at Johnston Atoll during cruise HA-15-01. All of these REA sites were selected using a stratified random design.

Table B.3.1.--Summary of sites where REA fish surveys were conducted at Johnston Atoll during cruise HA-15-01.

REA Site	Date	Depth Bin	Reef Zone	Depth (m)	Latitude	Longitude
JOH-495	26-Jan-15	Moderate	Forereef	12.4	16.69	-169.55
JOH-498	26-Jan-15	Deep	Forereef	22	16.76	-169.53
JOH-499	26-Jan-15	Deep	Forereef	18.8	16.74	-169.55
JOH-516	26-Jan-15	Moderate	Forereef	11.7	16.78	-169.50
JOH-517	26-Jan-15	Moderate	Forereef	11.6	16.75	-169.54
JOH-519	26-Jan-15	Moderate	Forereef	15.5	16.77	-169.52
JOH-520	26-Jan-15	Moderate	Forereef	10.9	16.70	-169.56
JOH-494	27-Jan-15	Moderate	Forereef	14.8	16.71	-169.49
JOH-496	27-Jan-15	Moderate	Forereef	14.9	16.71	-169.51

JOH-525	27-Jan-15	Deep	Lagoon	19.5	16.71	-169.52
JOH-536	27-Jan-15	Moderate	Lagoon	9	16.72	-169.48
JOH-539	27-Jan-15	Moderate	Lagoon	17	16.71	-169.52
JOH-552	27-Jan-15	Shallow	Lagoon	5.5	16.73	-169.50
JOH-579	27-Jan-15	Deep	Forereef	20.7	16.69	-169.53
JOH-581	27-Jan-15	Deep	Forereef	21.2	16.67	-169.55
JOH-478	28-Jan-15	Deep	Forereef	23	16.71	-169.40
JOH-483	28-Jan-15	Moderate	Forereef	14.3	16.73	-169.43
JOH-500	28-Jan-15	Moderate	Forereef	9.7	16.72	-169.46
JOH-543	28-Jan-15	Moderate	Lagoon	8.5	16.72	-169.47
JOH-547	28-Jan-15	Moderate	Lagoon	8	16.72	-169.49
JOH-561	28-Jan-15	Deep	Forereef	20.3	16.71	-169.45
JOH-570	28-Jan-15	Deep	Forereef	21.5	16.71	-169.43
JOH-594	28-Jan-15	Deep	Forereef	20	16.68	-169.48
JOH-484	29-Jan-15	Moderate	Forereef	11.5	16.74	-169.42
JOH-538	29-Jan-15	Moderate	Lagoon	12.7	16.74	-169.46
JOH-554	29-Jan-15	Shallow	Lagoon	6	16.74	-169.49
JOH-563	29-Jan-15	Deep	Forereef	26.1	16.72	-169.41
JOH-564	29-Jan-15	Deep	Forereef	23.3	16.70	-169.42
JOH-567	29-Jan-15	Deep	Forereef	30	16.71	-169.37
JOH-572	29-Jan-15	Deep	Forereef	20	16.74	-169.41
JOH-575	29-Jan-15	Deep	Forereef	21	16.69	-169.44

APPENDIX C: HOWLAND ISLAND

Howland Island is an uninhabited island located at 0°48′ N, 176°37′ W in the central Pacific and is part of the Pacific Remote Islands Marine National Monument. For information about the methods used to perform the activities discussed in this appendix, please see Appendix A: "Methods." In addition to the activities described in this appendix, a U.S. Fish and Wildlife Service field party went ashore to Howland Island during HA-15-01, Leg 1, to conduct surveys of terrestrial flora and fauna.

C.1. Ocean and Climate Change

Oceanographic operations during the cruise HA-15-01 at Howland Island entailed numerous retrievals and deployments of oceanographic moored instruments including the installation of subsurface temperature recorders (STRs) and calcification acidification units (CAUs), as well as nearshore water sampling and conductivity, temperature, and depth (CTD) casts and acoustic Doppler current profiler (ADCP) transects.

Six shallow-water CTD casts were performed at locations where water samples were collected. Nine shallow-water samples were collected for analysis of dissolved inorganic carbon (DIC), total alkalinity (TA), and salinity. In addition, 6 STRs and 22 CAUs were retrieved, and 6 STRs and 20 CAUs were deployed (Fig. C.1.1 and Table C.1.1).

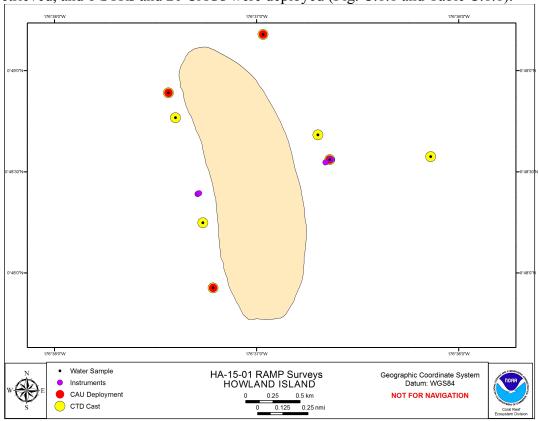


Figure C.1.1.--Mooring sites where oceanographic instruments and biological installations were retrieved or deployed and locations of nearshore CTD casts and water sampling performed at Howland Island during cruise HA-15-01.

Table C.1.1.- Geographic coordinates and depths of the moored oceanographic instruments (STRs) and biological installations (CAUs), that were retrieved or deployed at Howland Island during cruise HA-15-01.

		Instrument			Depth		
Site	Date	Type	Latitude	Longitude	(m)	Retrieved	Deployed
HOW_010	04-Feb-15	STR	0.80934	-176.61046	24.9	1	1
HOW_011	04-Feb-15	STR	0.80935	-176.61062	14.6	1	1
HOW_015	04-Feb-15	STR	0.80911	-176.61098	5.2	1	1
HOW_012	05-Feb-15	STR	0.80650	-176.62153	25.0	1	1
HOW_013	05-Feb-15	STR	0.80655	-176.62148	14.9	1	1
HOW_014	05-Feb-15	STR	0.80657	-176.62140	4.8	1	1
HOW-12	04-Feb-15	CAU	0.80935	-176.61063	15.2	5	5
HOW-05	05-Feb-15	CAU	0.80415	-176.62109	14.9	3	0
HOW-11	05-Feb-15	CAU	0.79877	-176.62025	13.7	5	5
HOW-13	06-Feb-15	CAU	0.81967	-176.61613	12.8	4	5
HOW-14	07-Feb-15	CAU	0.81485	-176.62393	14.6	5	5

C.2. Benthic and Microbial Communities

REA benthic survey sites were chosen using a stratified random design. Belt-transect surveys were conducted at 21 REA sites at Howland Island. Water samples for microbial analyses were collected at 3 sites (Fig. C.2.1; Table C.2.1). For more information about collections, see Table E.1.1 in Appendix E: "Biological Collections."

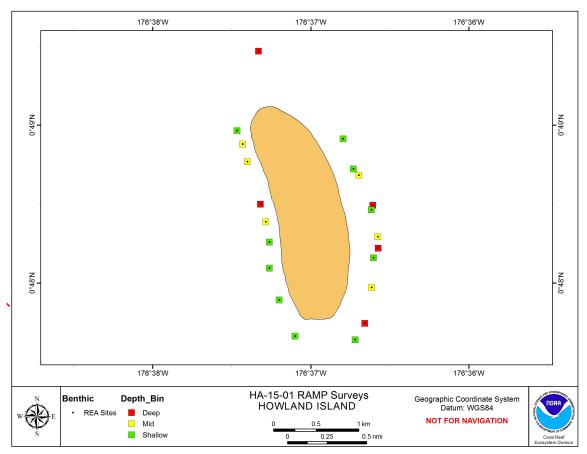


Figure C.2.1.--Locations of REA benthic sites surveyed at Howland Island during cruise HA-15-01. All of these REA sites were selected using a stratified random design.

Table C.2.1.—Summary of the REA benthic surveys and microbial water collections performed at Howland Island during cruise HA-15-01.

cruise IIA-13-		Depth		Depth			REA Coral	Microbial
REA Site	Date	Bin	Reef Zone	(ft)	Latitude	Longitude	Survey	Samples
HOW-12	04-Feb-15	Mid	Forereef	50	0.80935	-176.61063		Х
HOW-12	04-Feb-15	Mid	Forereef	82	0.80934	-176.61063		Х
HOW-604	04-Feb-15	Deep	Forereef	80	0.8082	-176.61012	Х	
HOW-655	04-Feb-15	Shallow	Forereef	21	0.81521	-176.61326	Х	
HOW-646	04-Feb-15	Mid	Forereef	49	0.81139	-176.6116	Х	
HOW-05	05-Feb-15	Mid	Forereef	82	0.8064	-176.62152		Х
HOW-611	05-Feb-15	Deep	Forereef	72	0.79574	-176.61097	Х	
HOW-05	05-Feb-15	Mid	Forereef	49	0.80415	-176.62109		Х
HOW-617	05-Feb-15	Deep	Forereef	78	0.80368	-176.60955	Х	
HOW-694	05-Feb-15	Shallow	Forereef	21	0.80774	-176.61028	Х	
HOW-690	05-Feb-15	Shallow	Forereef	16	0.79401	-176.61198	Х	
HOW-648	05-Feb-15	Mid	Forereef	47	0.79953	-176.61024	Х	
HOW-636	05-Feb-15	Mid	Forereef	45	0.81467	-176.62386	Х	
HOW-688	05-Feb-15	Shallow	Forereef	17	0.80158	-176.62103	Х	
HOW-697	05-Feb-15	Shallow	Forereef	20	0.80432	-176.62103	Х	
HOW-672	06-Feb-15	Shallow	Forereef	14	0.80267	-176.61004	Х	
HOW-607	06-Feb-15	Deep	Forereef	85	0.82447	-176.62218	х	
HOW-650	06-Feb-15	Shallow	Forereef	22	0.79821	-176.62	Х	
HOW-600	06-Feb-15	Deep	Forereef	74	0.80832	-176.62197	Х	
HOW-640	06-Feb-15	Mid	Forereef	46	0.8049	-176.60959	Х	
HOW-647	06-Feb-15	Mid	Forereef	51	0.8128	-176.62335	Х	
HOW-642	06-Feb-15	Mid	Forereef	48	0.80647	-176.62143	Х	
HOW-14	07-Feb-15	Mid	Forereef	51	0.8064	-176.62152		Х
HOW-687	07-Feb-15	Shallow	Forereef	21	0.81205	-176.61218	Х	
HOW-699	07-Feb-15	Shallow	Forereef	20	0.7944	-176.61833	х	
HOW-14	07-Feb-15	Mid	Forereef	48	0.81485	-176.62393		Х
HOW-686	07-Feb-15	Shallow	Forereef	20	0.81608	-176.62444	Х	

Additionally, during the HA-15-01 cruise, 6 towed-diver surveys were completed around Howland Island, covering a total length of 11.2 km of the ocean floor (Fig. C.2.2).



Figure C.2.2.--Track locations of towed-diver surveys conducted at Howland Island during the cruise HA-15-01.

C.3. Reef Fish Communities

REA fish survey sites were chosen using a stratified random design. Stationary-point-count surveys were conducted at 35 REA sites at Howland Island over three different habitat strata: deep, moderate, and shallow forereef (Fig. C.3.1 and Table C.3.1). No fishes were collected during these surveys.

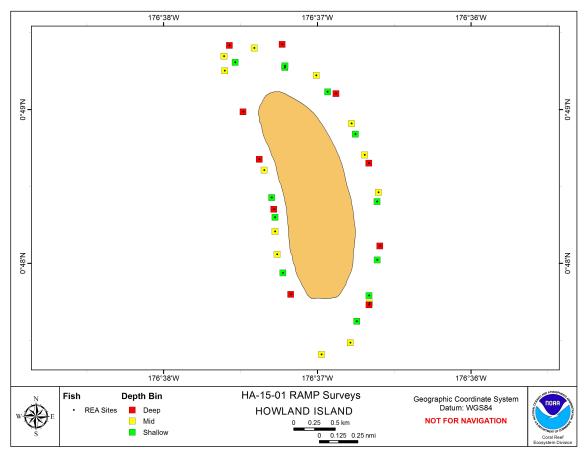


Figure C.3.1.-Locations of REA fish sites surveyed at Howland Island during cruise HA-15-01. All of these REA sites were selected using a stratified random design.

Table C.3.1.--Summary of sites where REA fish surveys were conducted at Howland Island during cruise HA-15-01.

1111 13 01.						
				Depth		
REA Site	Date	Depth Bin	Reef Zone	(m)	Latitude	Longitude
HOW-443	03-Feb-15	Moderate	Forereef	15	0.81	-176.62
HOW-457	03-Feb-15	Deep	Forereef	18	0.81	-176.62
HOW-440	04-Feb-15	Moderate	Forereef	13	0.82	-176.62
HOW-451	04-Feb-15	Deep	Forereef	22	0.82	-176.61
HOW-461	04-Feb-15	Moderate	Forereef	13.4	0.81	-176.61
HOW-465	04-Feb-15	Shallow	Forereef	5.1	0.81	-176.61
HOW-466	04-Feb-15	Deep	Forereef	18.2	0.80	-176.61
HOW-474	04-Feb-15	Shallow	Forereef	5	0.82	-176.62

HOW-486	04-Feb-15	Shallow	Forereef	6.1	0.81	-176.61
HOW-494	04-Feb-15	Shallow	Forereef	6	0.82	-176.63
HOW-437	05-Feb-15	Moderate	Forereef	12	0.79	-176.61
HOW-447	05-Feb-15	Shallow	Forereef	7	0.80	-176.62
HOW-448	05-Feb-15	Deep	Forereef	20.2	0.80	-176.62
HOW-458	05-Feb-15	Moderate	Forereef	10.9	0.82	-176.61
HOW-463	05-Feb-15	Deep	Forereef	23.3	0.82	-176.62
HOW-464	05-Feb-15	Moderate	Forereef	13.5	0.80	-176.61
HOW-467	05-Feb-15	Moderate	Forereef	13	0.82	-176.63
HOW-489	05-Feb-15	Shallow	Forereef	5.5	0.82	-176.62
HOW-497	05-Feb-15	Moderate	Forereef	15.8	0.82	-176.63
HOW-433	06-Feb-15	Deep	Forereef	21	0.82	-176.62
HOW-438	06-Feb-15	Shallow	Forereef	5.7	0.82	-176.62
HOW-452	06-Feb-15	Moderate	Forereef	13	0.81	-176.61
HOW-453	06-Feb-15	Shallow	Forereef	4	0.79	-176.61
HOW-455	06-Feb-15	Moderate	Forereef	13.9	0.80	-176.62
HOW-469	06-Feb-15	Deep	Forereef	24.5	0.80	-176.61
HOW-480	06-Feb-15	Shallow	Forereef	6	0.81	-176.62
HOW-506	06-Feb-15	Shallow	Forereef	5.5	0.80	-176.61
HOW-532	06-Feb-15	Deep	Forereef	20.6	0.81	-176.62
HOW-431	07-Feb-15	Moderate	Forereef	14.2	0.82	-176.62
HOW-449	07-Feb-15	Moderate	Forereef	12.4	0.80	-176.62
HOW-473	07-Feb-15	Deep	Forereef	26	0.81	-176.61
HOW-477	07-Feb-15	Shallow	Forereef	5.2	0.80	-176.62
HOW-490	07-Feb-15	Deep	Forereef	22.1	0.82	-176.63
HOW-493	07-Feb-15	Moderate	Forereef	13.5	0.79	-176.62
HOW-500	07-Feb-15	Shallow	Forereef	5.5	0.80	-176.61

APPENDIX D: BAKER ISLAND

Baker Island is an uninhabited island located at 0°12′ N, 176°29′ W in the central Pacific and is part of the Pacific Remote Islands Marine National Monument. For information about the methods used to perform the activities discussed in this appendix, please see Appendix A: "Methods." In addition to the activities described in this appendix, a U.S. Fish and Wildlife Service field party went ashore to Baker Island during HA-15-01, Leg 1, to conduct surveys of terrestrial flora and fauna.

D.1. Ocean and Climate Change

Oceanographic operations during the cruise HA-15-01 at Baker Island entailed numerous retrievals and deployments of oceanographic moored instruments including the installation of subsurface temperature recorders (STRs) and calcification acidification units (CAUs), as well as nearshore water sampling and conductivity, temperature, and depth (CTD) casts, and acoustic Doppler current profiler (ADCP) transects.

Eight shallow-water CTD casts were performed at locations where water samples were collected. Eight shallow-water samples were collected for analysis of dissolved inorganic carbon (DIC), total alkalinity (TA), and salinity. In addition, 7 STRs and 15 CAUs were retrieved, and 7 STRs and 20 CAUs were deployed (Fig. D.1.1 and Table D.1.1).

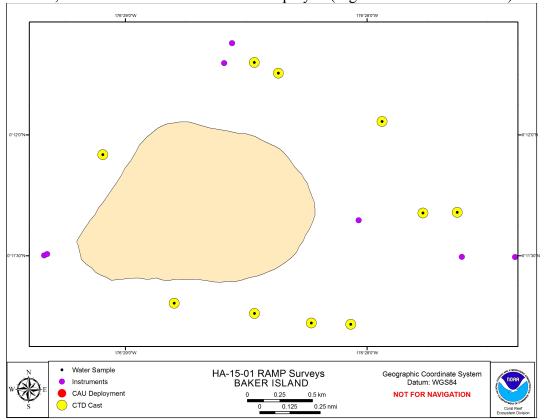


Figure D.1.1.--Mooring sites where oceanographic instruments and biological installations were retrieved or deployed and locations of nearshore CTD casts and water sampling performed at Baker Island during cruise HA-15-01.

Table D.1.1.- Geographic coordinates and depths of the moored oceanographic instruments (STRs) and biological installations (CAUs), that were retrieved or deployed at Baker Island during cruise HA-15-01.

		/,		<u> </u>			
		Instrument			Depth		
Site	Date	Type	Latitude	Longitude	(m)	Retrieved	Deployed
BAK_015	08-Feb-15	STR	0.20495	-176.47652	13.7	1	1
BAK_016	08-Feb-15	STR	0.19168	-176.48893	27.7	1	1
BAK_017	08-Feb-15	STR	0.19178	-176.48874	15.2	1	1
BAK_011	09-Feb-15	STR	0.19159	-176.46012	14.6	1	1
BAK_012	09-Feb-15	STR	0.19411	-176.46723	4.9	1	1
BAK_013	09-Feb-15	STR	0.20633	-176.47597	25.6	1	1
BAK_010	10-Feb-15	STR	0.19157	-176.45644	25.3	1	1
BAK-11	08-Feb-15	CAU	0.19864	-176.48489	14.3	3	5
BAK-14	10-Feb-15	CAU	0.20500	-176.47444	15.2	3	5
BAK-16	10-Feb-15	CAU	0.19461	-176.46280	12.2	2	0
BAK-02	11-Feb-15	CAU	0.18839	-176.47997	15.2	5	5
BAK-09	11-Feb-15	CAU	0.18678	-176.47000	19.2	2	0
BAK-50	12-Feb-15	CAU	0.18702	-176.47050	14.9	0	5

D.2. Benthic and Microbial Communities

REA benthic survey sites were chosen using a stratified random design. Belt-transect surveys were conducted at 15 REA sites at Baker Island. Water samples for microbial analyses were collected at 6 sites (Fig. D.2.1; Table D.2.1). For more information about collections, see Table E.1.1 in Appendix E: "Biological Collections."

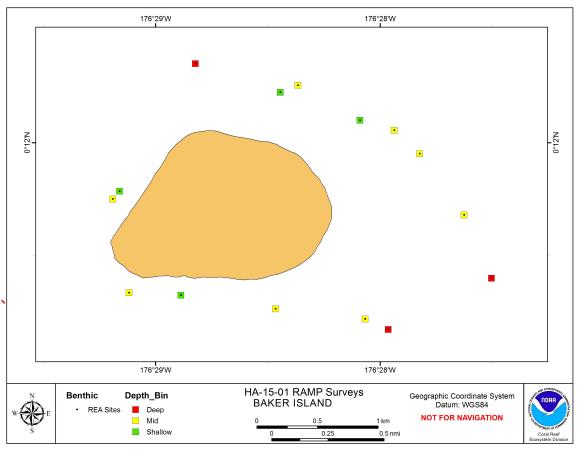


Figure D.2.1.-Locations of REA benthic sites surveyed at Baker Island during cruise HA-15-01. All of these REA sites were selected using a stratified random design.

Table D.2.1.--Summary of the REA benthic surveys and microbial water collections performed at Baker Island during cruise HA-15-01.

		Depth	Reef				REA Coral	Microbial
REA Site	Date	Bin	Zone	Depth (ft)	Latitude	Longitude	Survey	Samples
BAK-1999	08-Feb-15			50	0.19178	-176.48874		Х
BAK-11	08-Feb-15	Mid	Forereef	47	0.19864	-176.48489		Х
BAK-497	08-Feb-15	Shallow	Forereef	21	0.20374	-176.47408	Х	
BAK-480	08-Feb-15	Shallow	Forereef	21	0.20167	-176.46816	Х	
BAK-506	08-Feb-15	Shallow	Forereef	16	0.1887	-176.48146	Х	
BAK-1998	09-Feb-15			84	0.20633	-176.47597		Х

BAK-11	09-Feb-15	Mid	Forereef	48	0.19864	-176.48489		Х
BAK-408	09-Feb-15	Deep	Forereef	74	0.20587	-176.48038	Х	
BAK-457	09-Feb-15	Mid	Forereef	47	0.20426	-176.47277	Х	
BAK-454	09-Feb-15	Mid	Forereef	45	0.19583	-176.48652	Х	
BAK-490	09-Feb-15	Shallow	Forereef	20	0.19642	-176.48601	Х	
BAK-14	10-Feb-15	Mid	Forereef	50	0.205	-176.47444		Х
BAK-16	10-Feb-15	Mid	Forereef	40	0.19461	-176.4628		Х
BAK-407	10-Feb-15	Deep	Forereef	69	0.18996	-176.45839	Х	
BAK-418	10-Feb-15	Deep	Forereef	69	0.18616	-176.46606	Х	
BAK-452	10-Feb-15	Mid	Forereef	48	0.19465	-176.46043	Х	
BAK-462	10-Feb-15	Mid	Forereef	49	0.18693	-176.46777	Х	
BAK-02	11-Feb-15	Mid	Forereef	50	0.18839	-176.47997		Х
BAK-469	11-Feb-15	Mid	Forereef	48	0.18768	-176.47443	Х	
BAK-441	11-Feb-15	Mid	Forereef	38	0.20092	-176.46562	Х	
BAK-467	11-Feb-15	Mid	Forereef	49	0.19919	-176.46373	Х	
BAK-445	11-Feb-15	Mid	Forereef	49	0.18888	-176.48531	Х	

Additionally, during the HA-15-01 cruise, 5 towed-diver surveys were completed around Baker Island, covering a total length of 11.5 km of the ocean floor (Fig. D.2.2).

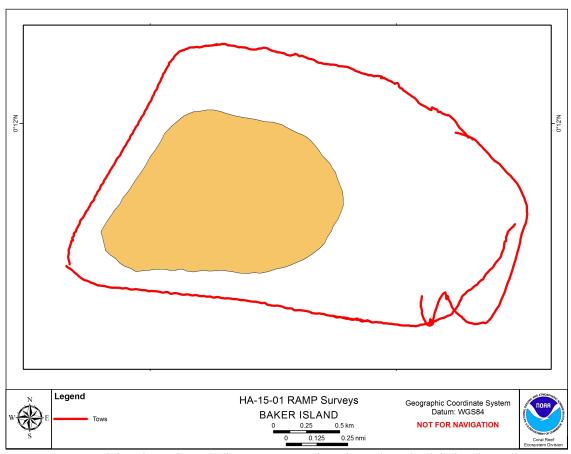


Figure D.2.2.-Track locations of towed-diver surveys conducted at Baker Island during the cruise HA-15-01.

D.3. Reef Fish Communities

REA fish survey sites were chosen using a stratified random design. Stationary-point-count surveys were conducted at 36 REA sites at Baker Island over three different habitat strata: deep, moderate, and shallow forereef (Fig. D.3.1 and Table D.3.1). No fishes were collected during these surveys.

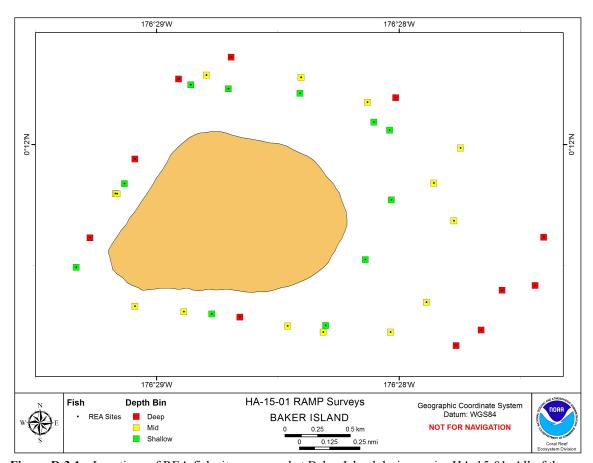


Figure D.3.1.--Locations of REA fish sites surveyed at Baker Island during cruise HA-15-01. All of these REA sites were selected using a stratified random design.

Table D.3.1.--Summary of sites where REA fish surveys were conducted at Baker Island during cruise HA-15-01.

REA_Site	Date	Depth Bin	Reef Zone	Depth (m)	Latitude	Longitude
BAK-247	08-Feb-15	Deep	Forereef	23	0.19	-176.48
BAK-260	08-Feb-15	Deep	Forereef	23	0.19	-176.46
BAK-273	08-Feb-15	Moderate	Forereef	15.5	0.19	-176.48
BAK-287	08-Feb-15	Moderate	Forereef	14.5	0.19	-176.47
BAK-307	08-Feb-15	Moderate	Forereef	12	0.19	-176.46
BAK-331	08-Feb-15	Shallow	Forereef	4.8	0.20	-176.47
BAK-333	08-Feb-15	Shallow	Forereef	5.5	0.19	-176.49
BAK-335	08-Feb-15	Shallow	Forereef	5	0.19	-176.47
BAK-336	08-Feb-15	Shallow	Forereef	5.4	0.20	-176.47

BAK-350	08-Feb-15	Shallow	Forereef	5.2	0.20	-176.49
BAK-254	09-Feb-15	Deep	Forereef	22.2	0.20	-176.47
BAK-261	09-Feb-15	Deep	Forereef	22.7	0.21	-176.48
BAK-302	09-Feb-15	Moderate	Forereef	16.7	0.20	-176.47
BAK-308	09-Feb-15	Moderate	Forereef	10	0.20	-176.47
BAK-313	09-Feb-15	Moderate	Forereef	15.2	0.20	-176.49
BAK-327	09-Feb-15	Shallow	Forereef	5.3	0.20	-176.48
BAK-328	09-Feb-15	Shallow	Forereef	5.2	0.20	-176.48
BAK-337	09-Feb-15	Shallow	Forereef	5.8	0.20	-176.47
BAK-357	09-Feb-15	Deep	Forereef	21.7	0.20	-176.48
BAK-243	10-Feb-15	Deep	Forereef	27	0.19	-176.46
BAK-257	10-Feb-15	Deep	Forereef	22	0.20	-176.48
BAK-284	10-Feb-15	Moderate	Forereef	15.8	0.19	-176.47
BAK-293	10-Feb-15	Moderate	Forereef	13.33	0.19	-176.47
BAK-295	10-Feb-15	Moderate	Forereef	13.4	0.19	-176.48
BAK-297	10-Feb-15	Moderate	Forereef	16.7	0.20	-176.46
BAK-298	10-Feb-15	Moderate	Forereef	10.1	0.19	-176.46
BAK-329	10-Feb-15	Shallow	Forereef	5	0.20	-176.47
BAK-360	10-Feb-15	Shallow	Forereef	4.1	0.19	-176.47
BAK-248	11-Feb-15	Deep	Forereef	22.7	0.19	-176.46
BAK-253	11-Feb-15	Deep	Forereef	21.2	0.19	-176.46
BAK-258	11-Feb-15	Deep	Forereef	19.2	0.19	-176.46
BAK-266	11-Feb-15	Deep	Forereef	20.2	0.19	-176.49
BAK-280	11-Feb-15	Moderate	Forereef	10.7	0.20	-176.48
BAK-282	11-Feb-15	Moderate	Forereef	8.1	0.20	-176.46
BAK-317	11-Feb-15	Shallow	Forereef	4.5	0.19	-176.48
BAK-320	11-Feb-15	Moderate	Forereef	11	0.20	-176.49

APPENDIX E: BIOLOGICAL COLLECTIONS

Biological and other samples were collected at Johnston, Baker, and Howland and their surrounding waters for multiple research purposes. These collections are listed here in Table E.1.1.

Table E.1.1.--Samples collected at Johnston, Baker, and Howland for microbial and ocean acidification

analyses during cruise HA-15-01.

<i>j</i>	ig cluise IIA-1.	7 01.							
				Specimen	Number of	Depth			
REA Site	Date	Latitude	Longitude	Collected	Samples	(m)			
	Microbial Collections: Water Samples, Coral Rubble, and Macroalgae								
JOH-11	26-Jan-15	16.7215	-169.52412	2 L	2	11.6			
JOH-1999	27-Jan-15	16.67068	-169.52682	2 L	2	25			
JOH-56	27-Jan-15	16.691667	-169.52699	2 L	2	13.7			
JOH-56	27-Jan-15	16.691667	-169.52699	20 L	3	13.7			
JOH-09	28-Jan-15	16.72856	-169.5119	2 L	2	7			
JOH-10	28-Jan-15	16.76343	-169.5119	2 L	2	14.6			
JOH-10	28-Jan-15	16.76343	-169.5119	20 L	3	14.6			
JOH-12	29-Jan-15	16.741034	-169.524569	2 L	2	11			
JOH-12	29-Jan-15	16.741034	-169.524569	20 L	3	11			
HOW-12	04-Feb-15	0.80934	-176.61063	2 L	2	25			
HOW-12	04-Feb-15	0.80935	-176.61063	2 L	2	13.7			
HOW-12	04-Feb-15	0.80935	-176.61063	20 L	3	13.7			
HOW-05	05-Feb-15	0.80415	-176.62109	2 L	2	14.3			
HOW-13	06-Feb-15	0.81967	-176.61613	2 L	2	12.8			
HOW-13	06-Feb-15	0.81967	-176.61613	20 L	3	12.8			
BAK-11	08-Feb-15	0.19864	-176.48489	2 L	2	14.3			
BAK-1999	08-Feb-15	0.19178	-176.48874	2 L	2	15.2			
BAK-11	09-Feb-15	0.19864	-176.48489	2 L	2	14.3			
BAK-11	09-Feb-15	0.19864	-176.48489	20 L	3	14.3			
BAK-1998	09-Feb-15	0.20633	-176.47597	2 L	2	25.3			
BAK-14	10-Feb-15	0.205	-176.47444	2 L	2	14.9			
BAK-16	10-Feb-15	0.19461	-176.4628	2 L	2	11.9			
BAK-02	11-Feb-15	0.18839	-176.47997	2 L	2	14.9			
BAK-02	11-Feb-15	0.18839	-176.47997	20 L	3	14.9			
Algal Collec	tions: Ocean A	Acidification							
JOH-11	26-Jan-15	16.72	-169.52	CAU UNIT	5	11.8872			
JOH-12	27-Jan-15	16.75	-169.52	CAU UNIT	5	11.5824			
JOH-07	28-Jan-15	16.71	-169.48	CAU UNIT	5	12.192			
JOH-10	28-Jan-15	16.76	-169.51	CAU UNIT	5	14.6304			
JOH-09	28-Jan-15	16.73	-169.49	CAU UNIT	5	7.62			
HOW-12	04-Feb-15	0.81	-176.61	CAU UNIT	5	15.24			
HOW-11	05-Feb-15	0.80	-176.62	CAU UNIT	5	13.716			
HOW-05	05-Feb-15	0.80	-176.62	CAU UNIT	3	14.9352			

HOW-13	06-Feb-15	0.82	-176.62	CAU UNIT	4	12.8016
HOW-14	07-Feb-15	0.81	-176.62	CAU UNIT	5	14.6304
BAK-11	08-Feb-15	0.20	-176.48	CAU UNIT	3	15.24
BAK-16	10-Feb-15	0.19	-176.46	CAU UNIT	2	12.192
BAK-14	10-Feb-15	0.21	-176.47	CAU UNIT	3	15.24
BAK-02	11-Feb-15	0.19	-176.48	CAU UNIT	5	14.6304
BAK-09	11-Feb-15	0.19	-176.47	CAU UNIT	2	19.2024