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## CRUISE REPORT<sup>1</sup>

**VESSEL:** NOAA Ship *Hi`ialakai*, Cruise HA-11-01, Leg I

**CRUISE PERIOD:** 10 March–2 April 2011

**AREA OF OPERATION:** Wake Atoll

**TYPE OF OPERATION:** Personnel from the Coral Reef Ecosystem Division (CRED) of the NOAA Pacific Islands Fisheries Science Center (PIFSC), San Diego State University (SDSU), and Papahānaumokuākea Marine National Monument (PMNM) conducted interdisciplinary Pacific Reef and Assessment Monitoring Program (Pacific RAMP) surveys in waters surrounding Wake Atoll, part of the Pacific Remote Islands Marine National Monument (PRIMNM). A summary of Pacific RAMP planned operations were sent to the U.S. Fish and Wildlife Service (USFWS), PRIMNM, and U.S. Air Force (USAF) base command at Wake Atoll prior to the onset of cruise HA-11-01, Leg I, although no permits were required.

The necessary evacuations of personnel from Laysan Island and Kure Atoll resulted in the unplanned entry into the PMNM. These evacuation activities did not require a permit from PMNM managers, per the final regulations that codify the prohibitions and management measures set forth in presidential proclamation 8031, which established the PMNM. Specifically, Title 50, Part 404.8 of the *Code of Federal Regulations* stipulates that PMNM “prohibitions in this part do not apply to activities necessary to respond to emergencies threatening life, property, or the environment, or to activities necessary for law enforcement purposes.”

Prior to the *Hi`ialakai* entering this management area, the NOAA permit coordinator for the PMNM was notified of our intentions and circumstances. Additionally, the ship was in compliance with the resource protection measures typically required by permits, including a clean hull, the implementation of proper ballast water and discharge procedures, a vessel free of rodents, and a Vessel Monitoring System in place.

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<sup>1</sup> PIFSC Cruise Report CR-11-004  
Issued 13 May 2011



## ITINERARY:

Note: Daily field operations included Rapid Ecological Assessment (REA) benthic surveys, REA fish surveys, and towed-diver surveys of both benthic and fish communities. Unless otherwise specified in the following daily summaries, these surveys occurred during each operational day.

- 10 March Start of cruise. Embarked all scientific crew. After fueling was complete, the *Hi`ialakai* departed Pearl Harbor, Honolulu, O`ahu Island, at 1200 and began transit to Wake Atoll.
- 11 March Transit day. Diverted course at 1500 to Laysan Island to evacuate PIFSC and USFWS personnel impacted by the tsunami generated by the Japan earthquake.
- 12 March Transit day.
- 13 March Transit day.
- 14 March Arrived at Laysan Island for evacuation of personnel. Embarked PIFSC personnel Erin Prickett, Arthur Wong, and Adam Fox and USFWS personnel John Watson, Andrea Kristof, Pam Tyhurst, and Emily Cook. Began transit to Midway Atoll.
- 15 March Arrived at Midway Atoll for transfer of PIFSC and USFWS evacuees. Disembarked PIFSC personnel Erin Prickett, Arthur Wong, and Adam Fox and USFWS personnel John Watson, Andrea Kristof, Pam Tyhurst, and Emily Cook. Began transit to Kure Atoll.
- 16 March Arrived at Kure Atoll for evacuation of island personnel. Embarked PIFSC personnel Jason Jones and State of Hawai`i Department of Land and Natural Resources (DLNR) personnel Cynthia Vanderlip, Julie Parish, Cynthia Waddington, and Jim Waddington. Retrieved one deepwater (125 m) ecological acoustic recorder (EAR) that was located approximately 11.3 km to the southeast of Kure Atoll at a depth of 125 m. Began transit to Midway Atoll.
- 17 March Arrived at Midway Atoll for transfer of PIFSC and DLNR evacuees. Disembarked PIFSC personnel Jason Jones and DLNR personnel Cynthia Vanderlip, Julie Parish, Cynthia Waddington, and Jim Waddington. Began transit to Wake Atoll.
- 19 March Transit day. Crossed dateline: March 18 became March 19.
- 20 March Transit day.

- 21 March Transit day.
- 22 March Transit day.
- 23 March Arrived at Wake Atoll and began field operations. Deployed and retrieved the following types of instruments: subsurface temperature recorder (STR), remote access sampler (RAS), conductivity, temperature, depth (CTD) sensor, temperature and salinity sensor (SBE 37 MicroCAT, Sea-Bird Electronics Inc., Bellevue, Wash.), acoustic Doppler profiler (ADP), autonomous reef monitoring structure (ARMS), and calcification acidification unit (CAU). Nearshore water samples were collected for chlorophyll-*a* (Chl-*a*), nutrient, dissolved inorganic carbon (DIC), total alkalinity (TA) salinity, and microbial community analyses. Nearshore CTD profiles were collected, and shipboard operations included acoustic Doppler current profiler (ADCP) transects, deepwater CTD casts, and water sampling for Chl-*a* and nutrient concentrations. The 17-ft Avon boat was deployed and moored in the small-boat harbor for later use.
- 24 March Continued field operations at Wake Atoll. Deployed and retrieved the following types of instruments: STR, SBE 37, ADP, ARMS, and CAU. Algal voucher specimens were collected for taxonomic identification. Nearshore water samples were collected for Chl-*a*, nutrient, DIC, TA, salinity, and microbial community analyses. Nearshore CTD profiles were collected, and shipboard operations included ADCP transects and deepwater CTD casts.
- 25 March Continued field operations at Wake Atoll. Oceanography team entered the lagoon for operations and used the 17-ft Avon. Deployed and retrieved the following types of instruments: sea-surface temperature (SST) buoy, STR, SBE 37, ARMS, CAU, and EAR. Algal voucher specimens were collected for taxonomic identification. Nearshore water samples were collected for Chl-*a*, nutrient, DIC, TA, salinity, and microbial community analyses. Nearshore CTD profiles were collected, and shipboard operations included ADCP transects and deepwater CTD casts and water sampling for Chl-*a* and nutrient concentrations. A high-frequency acoustic recording package (HARP) was recovered and deployed at a depth of 800 m approximately 4.8 km southeast of Wake Atoll.
- 26 March Continued field operations at Wake Atoll. ARMS team entered the lagoon for operations and used the 17-ft Avon. Retrieved and deployed the following types of instruments: STR, RAS, CTD, ADP, SBE 37, CAU, and ARMS. Algal voucher specimens were collected for taxonomic identification, and samples of the algal genus *Halimeda* were collected for calcification analysis. Nearshore water samples were collected for Chl-*a*, nutrient, DIC, TA, salinity, and microbial community analyses. Nearshore

CTD profiles were collected, and shipboard operations included ADCP transects and deepwater CTD casts.

- 27 March Continued field operations at Wake Atoll. Coral cores were collected from REA benthic sites WAK-08 and WAK-05. Towed-diver team conducted fish and benthic calibration tows to compare interobserver variability. Recovered 17-ft Avon. Began transit to Saipan Island.
- 28 March Transit day.
- 29 March Transit day.
- 30 March Transit day.
- 31 March Transit day.
- 1 April Transit day.
- 2 April Arrived at Saipan Island, part of the Commonwealth of the Northern Mariana Islands (CNMI). Recovered 1 HARP located at a depth of 1200 m and approximately 27.4 km northwest of Saipan. Pulled into Saipan Harbor. Deployed two 19-ft boats (SAFE Boats International) and delivered them to the small-boat harbor. Off-loaded retired Coral Reef Early Warning System (CREWS) buoys transported at the request of the CNMI Division of Fish and Wildlife (DFW). End of cruise HA-11-01, Leg I.

## **MISSIONS:**

- A. Conducted ecosystem monitoring of the species composition, abundance, percentage of cover, size distribution, and general health of the fishes, corals, target macroinvertebrates, and algae of the shallow-water ( $\leq 30$  m) coral reef ecosystems of Wake Atoll.
- B. Deployed and retrieved an array of instruments and installations—including SST buoys, STRs, ADPs, temperature and salinity sensors, CTD sensors, ARMS, CAUs, and EARs—to allow for remote, long-term monitoring of oceanographic, environmental, and ecological conditions of the coral reef ecosystems of Wake Atoll.
- C. Conducted shallow-water CTD casts and collected water samples for Chl-*a*, nutrient, DIC, TA, salinity, and microbial community analyses to depths  $\leq 30$  m to examine physical and biological linkages supporting and maintaining these island ecosystems.
- D. Conducted shipboard oceanographic and meteorological observations to examine physical and biological linkages supporting and maintaining these island ecosystems, using CTD casts deployed to a depth of 500 m with concurrent water samples taken at select locations and depths, collecting continuous ADCP, SST, salinity, and partial pressure of carbon dioxide (pCO<sub>2</sub>) data around reef ecosystems and fundamental meteorological data, such as air temperature, wind speed and direction, barometric pressure, and relative humidity.
- E. Collected a small number of shallow-water coral cores to examine calcification (growth) rates in recent decades and assess potential early impacts of ocean acidification.
- F. Determined the existence of threats to the health of these coral reef resources from anthropogenic sources, including marine debris.
- G. Deployed and retrieved HARPs at Wake Atoll and retrieved HARP at Saipan Island. Part of the cetacean research program headed by Erin Oleson, PhD, of the PIFSC Protected Species Division, the HARP passively records ambient underwater sounds produced by biological, environmental, and man-made sources.

## **RESULTS:**

This section provides tallies of research activities (Table 1), a summary of important observations, and a list of data collected during cruise HA-11-01, Leg I. For more information pertaining to the data collected, methodology employed, and preliminary findings at the islands visited, see Appendices A–C.

**Table 1.** Statistics for the Pacific RAMP 2011 cruise to Wake Atoll (cruise HA-11-01, Leg I), including activities at Kure Atoll and Saipan Island. The total numbers for REA sites include sites where REA benthic or fish surveys were conducted. The totals for scuba dives include all dives carried out for all activities at each island.

<b>Research Activity</b>	<b>Wake</b>	<b>Kure</b>	<b>Saipan</b>
Scuba Dives	243	0	0
<b>Biological Surveys</b>			
Towed-diver Surveys: Benthic and Fish	18	0	0
Combined Length (km) of Towed-diver Surveys	36.1	0	0
REA Sites: Benthic	11	0	0
REA Sites: Fish	30	0	0
<b>Biological Sample Collections</b>			
Algal Voucher Specimens (taxonomic ID)	6	0	0
<i>Halimeda</i> Samples (calcification analysis)	30	0	0
Coral Core Samples	2	0	0
Microbial Water Samples	20	0	0
Microbial Benthic Samples	23	0	0
<b>Biological Monitoring Installations</b>			
ARMS Retrieved	9	0	0
ARMS Deployed	9	0	0
CAUs Deployed	25	0	0
EAR Retrieved	1	0	0
Deepwater Ear Retrieved	0	1	0
HARP Retrieved	1	0	1
HARP Deployed	1	0	0
<b>Oceanographic Moored Instruments</b>			
SST Buoy Retrieved	1	0	0
SST Buoy Deployed	1	0	0
STRs Retrieved	11	0	0
STRs Deployed	11	0	0
ADPs Retrieved	3	0	0
ADP Deployed	1	0	0
CTD Sensor Retrieved	1	0	0
CTD Sensor Deployed	1	0	0
Temperature and Salinity Sensors (SBE 37) Retrieved	6	0	0
<b>Hydrographic Surveys</b>			
Shallow-water CTD Casts	5	0	0
Deepwater CTD Casts	31	0	0
Total Length (km) of ADCP Transects	100	0	0
<b>Water-quality Sampling</b>			
Shallow-water Chl- <i>a</i> Water Samples	10	0	0
Shallow-water Nutrient Water Samples	12	0	0
Shallow-water Salinity Water Samples	10	0	0
Shallow-water DIC Water Samples	10	0	0
Deepwater Chl- <i>a</i> Water Samples	10	0	0
Deepwater Nutrient Water Samples	10	0	0
DIC Samples via Shipboard pCO <sub>2</sub> System	8	0	0
RAS Deployed	1	0	0
RAS Retrieved	1	0	0

The coral reef ecosystem of Wake Atoll is surveyed biennially through CRED's Pacific RAMP. The cruise HA-11-01, Leg I, marked this program's fourth expedition to Wake Atoll. Here, we present highlights from our observations during this latest expedition.

## Wake Atoll

### *Reef Fishes*

- The bumphead parrotfish (*Bolbometopon muricatum*) was observed in small schools (~ 10–20 individuals) along the east coast or as individuals at REA sites on the southern and southeastern forereefs. Although not seen in large aggregations (200+), as was observed during the previous RAMP cruise in 2009, the bumphead parrotfish, which is listed as “vulnerable” by the International Union for Conservation of Nature (IUCN) and is a “species of concern” under the *Endangered Species Act*, was nonetheless seen during REA fish surveys far more commonly at Wake than at other islands and atolls surveyed by CRED.
- As in previous years, 1 or 2 humphead wrasses (*Cheilinus undulatus*) were observed during most fish surveys. Like the bumphead parrotfish, this rare fish is seen more often at Wake than at other CRED survey locations. It is listed by the IUCN as endangered.
- Small (40–100 cm) gray reef sharks (*Carcharhinus amblyrhynchos*) were seen in small schools (10–20 individuals) near the reef edge, between 10 and 20 m deep, on the southeastern and northwestern corners of this atoll, while larger individuals (120–150 cm) were seen in deeper waters (~20–30 m).
- Three new fish records for Wake Atoll were documented: the African pompano (*Alectis ciliaris*), midnight snapper (*Macolor macularis*), and black snapper (*Macolor niger*).

### *Oceanography*

- The oceanography team recovered numerous oceanographic instruments that measure ocean currents, temperature, salinity, and waves, enabling a detailed assessment of nearshore hydrodynamic conditions at Wake Atoll. These data are part of a multidisciplinary research project lead by Jamison Gove that is investigating the effects of physical forcing on benthic coral reef community structure.
- The oceanography team conducted the first investigative study of ocean carbonate chemistry and the effects of ocean acidification on reef ecosystems at Wake Atoll. This study included the deployment of the remote access sampler (RAS), which collected 48 water samples over a 2-day time period, providing data to assess variability in carbonate chemistry while constraining diel forcing, as well as the coring of lobed corals, providing historical coral accretion rates at Wake Atoll.

### *Benthic Environment*

- The shipwreck located southeast of the harbor mouth (REA site WAK-02) was surveyed and found to have minimal levels of cyanobacteria in the surrounding

- area. This finding was not expected, as it is common for the benthic substrate surrounding a shipwreck to have anomalously high levels of cyanobacteria.
- Coral disease appeared to have a low occurrence at Wake Atoll, compared to other reef environments visited by CRED.
  - A high percentage of cover of the brown macroalga *Lobophora spp.* was observed.

The following operations not related to RAMP were also associated with the cruise HA-11-01, Leg I:

#### Emergency response to Laysan Island and Kure Atoll

The Japan earthquake and subsequent tsunami generated on March 11, 2011, propagated across the Pacific, affecting many island locations, including Laysan Island and Kure Atoll, where PIFSC, USFWS, and DLNR personnel were stationed. Although none of the individuals sustained injuries, the food and water supplies on Laysan were compromised and the cooking shelter destroyed, resulting in their necessary evacuation. Personnel stationed at Kure Atoll suffered less direct impacts; however, the instability of Japan's nuclear facility combined with U.S. government budget uncertainties resulted in their necessary evacuation. The *Hi`ialakai* responded to aid these field stations, evacuating all island personnel and delivering them to Midway Atoll. Laysan Island personnel included Erin Prickett, Arthur Wong, and Adam Fox of PIFSC and John Watson, Andrea Kristof, Pam Tyhurst, and Emily Cook of USFWS. Kure Atoll personnel included Jason Jones of PIFSC and Cynthia Vanderlip, Julie Parish, Cynthia Waddington, and Jim Waddington of DLNR.

#### Kure Atoll

An EAR was opportunistically recovered on March 16 at approximately 1830 local time at 28°20'3" N, 178°15'12" W at a depth of 123 m approximately 11.3 km southeast of Kure Atoll. This EAR was deployed on May 21, 2010, by Anne Rosinski during a University of Hawai`i research cruise, as part of the bioacoustics research program headed by Marc Lammers, PhD, and Whitlow Au, PhD, at the Hawai`i Institute of Marine Biology (HIMB) of the University of Hawai`i. The EAR passively records ambient underwater sounds produced by biological, environmental, and man-made sources.

#### Wake Atoll and Saipan Island

Two HARPs were recovered and one redeployed during the cruise HA-11-01, Leg I: one retrieval and one deployment occurred on March 25 at 1400 at a depth of 800 m approximately 4.8 km southeast of Wake Atoll and one retrieval on April 2 at 0800 at a depth of 1200 m approximately 27.4 km northwest of Saipan Island.



The following data and samples were collected during this expedition:

**REA Benthic Surveys:**

- Digital still photographs of overall site character and typical benthos
- Digital still photographs of the benthos along transect lines
- Quantitative assessments of benthic composition from line-point-intercept surveys
- Algal voucher specimens necessary for algal species identification
- Samples of the algal genus *Halimeda* to determine degree of calcification for ocean acidification research
- Field notes of algal species diversity and relative abundance
- Number, species or genus, size, and health condition of all coral colonies observed within belt transects of known area
- Digital still photographs of diseased corals and coralline algae
- Field notes on signs of coral bleaching or disease
- Assessment of calcification rates from collected cores of massive reef building corals

**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, and habitat complexity
- Digital still photographs of the benthos along transect lines
- Digital still photographs of rare or interesting fish species
- Species presence checklists for estimates of fish community diversity

**Towed-diver Surveys:**

- Digital still photographs and video of benthic habitats
- Counts of target macroinvertebrates, including crown-of-thorns seastars, sea cucumbers, sea urchins, and giant clams
- Quantitative assessments of large ( $\geq 50$  cm in total length) reef fishes to species level
- Quantitative and qualitative assessments of key protected species and species of concern, including cetaceans, sea turtles, and rare fishes
- Benthic habitat characterization, including visual estimates of habitat complexity, habitat type, and cover of corals, stressed corals, macroalgae, and crustose coralline red algae
- Temperature and depth data

**Shipboard Oceanography:**

- Deepwater CTD profiles to a depth of 500 m
- Chl-*a* and nutrient concentrations from water samples collected at variable depths
- Dissolved oxygen, turbidity, and fluorescence measurements from CTD sensor
- Transects of profiles of ocean current velocity and direction collected using a shipboard ADCP unit
- Solar radiation, air temperature, barometric pressure, and wind speed and direction

- Select surface measurements of partial pressure of carbon dioxide (pCO<sub>2</sub>)
- Surface temperature and salinity measurements

**Nearshore Oceanography from Small Boats:**

- Shallow-water CTD profiles to depths  $\leq 30$  m at all REA sites where CAUs were installed, with dissolved oxygen measurements
- Concentrations of Chl-*a*, nutrients, salinity, DIC, and TA from water samples collected in concert with shallow-water ( $\leq 30$  m) CTD casts
- Temporary high-resolution carbonate chemistry from RAS deployments

**Biological Monitoring Installations:**

- Environmental acoustics of reefs, marine mammals, and boat traffic from EARs
- Assessment of taxonomic diversity of coral reef species by collection of invertebrate specimens from retrieved ARMS

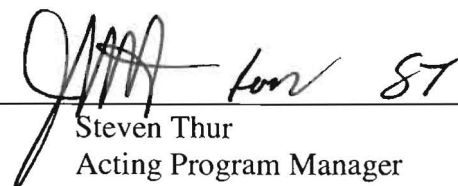
**Oceanographic Moored Instruments:**

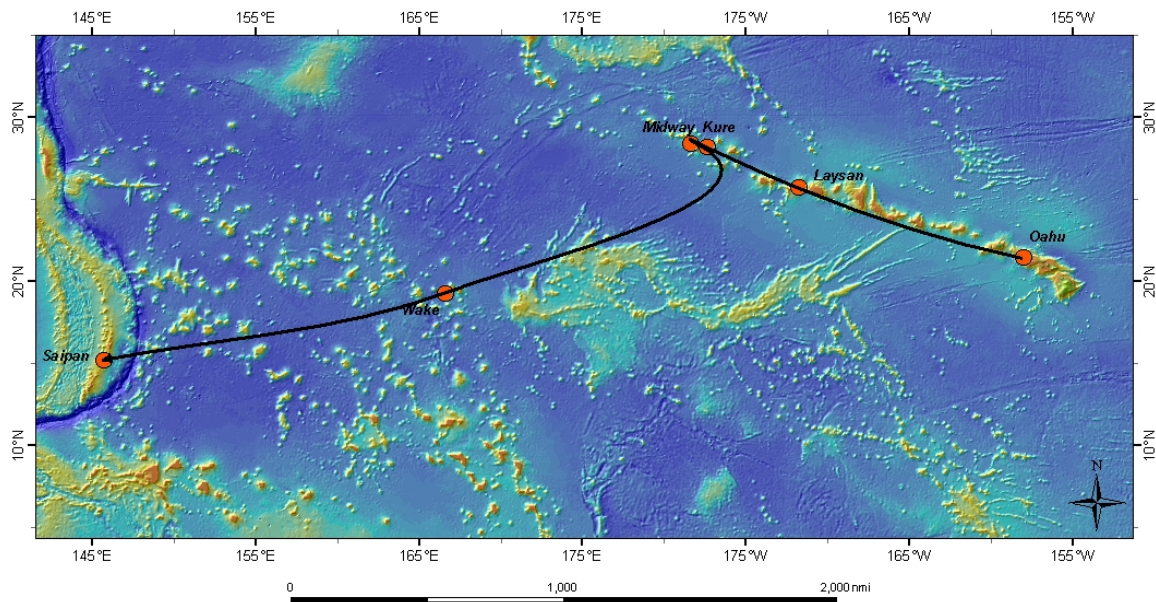
- Sea-surface and subsurface temperature at variable depths
- Subsurface salinity at variable depths
- ADP current profiles and wave spectra

**SCIENTIFIC PERSONNEL:**

Jamison Gove, Chief Scientist, Oceanography Team, University of Hawai'i (UH)-Joint Institute for Marine and Atmospheric Research (JIMAR), Pacific Islands Fisheries Science Center (PIFSC)-Coral Reef Ecosystems Division (CRED)  
Jesse Abdul, Data Manager, UH-JIMAR, PIFSC-CRED  
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Edmund Coccagna, Fish Team—Towed Diver, UH-JIMAR, PIFSC-CRED  
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Submitted by:   
Jamison Gove  
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Approved by:   
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**Figure 1.**--Track of the NOAA Ship *Hi'ialakai* for the cruise HA-11-01, Leg I, March 10–April 2, 2011, with Wake Atoll surveyed and Laysan Island and Kure Atoll visited because of tsunami evacuation response. Satellite image SIO, NOAA, U.S. Navy, NGA, GEBCO (Becker, 2009; Smith and Sandwell, 1997) © 2008 The Regents of the University of California.

## 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-11-01, Leg I, on the NOAA Ship *Hi'ialakai* during the period of 10 March–2 April 2011. The first Pacific RAMP expedition at Wake Atoll was conducted in 2005.

### A.1. Oceanography and Water Quality

*(Jamison Gove, Lisa Munger, Oliver Vetter, and Charles Young)*

To assess and monitor the oceanographic and water-quality parameters influencing the coral reef ecosystems at Wake Atoll, part of the Pacific Remote Islands Marine National Monument, the oceanography team performed the following activities: (1) conducted deepwater oceanographic surveys characterizing prevailing water properties and ocean currents around these islands, (2) completed nearshore oceanographic and water-quality surveys, and (3) deployed and retrieved an array of subsurface moored instruments designed to provide continuous, high-resolution time-series observations. Shipboard meteorological observations, including wind speed and direction, relative humidity, air temperature, and barometric pressure, were recorded. In addition, the oceanography team retrieved and deployed ecological acoustic recorders (EARs) and high-frequency acoustic recording packages (HARPs), both of which passively records ambient underwater sounds produced by biological, environmental, and man-made sources, and participated in installations of calcification acidification units (CAUs) for the assessment of calcification rates of crustose coralline red algae and hard corals (see Section A.2.3: “Installations for Monitoring Marine Life,” for information about CAU and EAR techniques).

#### 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 instrument platforms that internally record in situ observations and telemeter that data in near real time. The following types of oceanographic instruments were retrieved or deployed during this cruise.

**Sea-surface Temperature (SST) Buoy:** provides high-resolution SST (SBE 39 sensor, Sea-Bird Electronics Inc., Bellevue, Wash., accuracy of 0.002°C). Data are sampled at 30-min intervals and internally recorded. Subsets of these data are transmitted daily via satellite telemetry.

**Subsurface Temperature Recorder (STR):** provides near-real-time, high-resolution temperature data (SBE 39 sensor). Data are internally recorded at 30-min intervals. This type of subsurface instrument is deployed at depths of 0.5–40 m.

**Acoustic Doppler Profiler (ADP):** provides directional current profiles and wave spectra using a 3-beam-configured 1-MHz Aquadopp Profiler (Nortek, Rud, Norway, accuracy of accuracy of  $0.005 \text{ m s}^{-1}$  in current and 0.1% in pressure). Sample intervals for current and wave data vary depending on duration of deployment. This type of subsurface instrument is deployed at depths of 5–20 m.

**Remote Access Sampler (RAS):** the McLane Remote Access Sampler (East Falmouth, Mass.) is an autonomous water sampling instrument that can collect up to 48 water samples, each 500 mL, over a programmer-dictated time series. This instrument has the capability for high-frequency, hourly sampling. CRED uses this RAS in depths up to 30 m, but this sampler has a maximum sampling depth of 5500 m.

**Temperature and Salinity Sensor:** provides high-resolution temperature and conductivity data (SBE 37 MicroCAT). Conductivity data is used to calculate salinity.

### A.1.2. Hydrographic Surveys

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

**Shallow-water (Nearshore) Conductivity, Temperature, and Depth (CTD) Casts:** a CTD profiler deployed from a small boat provided data on temperature, conductivity, which is related to salinity, and pressure, which is related to depth (SBE 19*plus* Seacat Profiler, accuracy of  $0.005 \text{ S m}^{-1}$  in conductivity,  $0.0002^\circ\text{C}$  in temperature, and 0.1% in pressure). 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) was also attached, and measurements were made in concert with CTD measurements. A CTD cast was performed at each of the Rapid Ecological Assessment (REA) sites where CAUs were deployed. Data were collected by hand lowering this profiler off a small boat at descent rates of  $\sim 0.5\text{--}0.75 \text{ m s}^{-1}$  at depths  $\leq 30 \text{ m}$ .

**Deepwater (Shipboard) CTD Casts:** a ship-based CTD profiler provided high-resolution conductivity, temperature, and pressure data (SBE 911*plus* CTD, accuracy of  $0.003 \text{ S m}^{-1}$  in conductivity,  $0.001^\circ\text{C}$  in temperature, and 0.015% in pressure). Measurements of dissolved oxygen (SBE43) and fluorescence and turbidity (*ECO* FLNTU, WET Labs, accuracy of  $0.01 \mu\text{g l}^{-1}$  in fluorescence and 0.01 NTU in turbidity) were performed in concert with CTD measurements. Data were collected at depths up to 500 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. Data were continuously collected throughout the research cruise.

**Water Chemistry:** water samples for analyses of concentrations of chlorophyll-*a* (Chl-*a*), dissolved inorganic carbon (DIC), total alkalinity (TA), and nutrients—nitrate, NO<sub>3</sub><sup>-</sup>; nitrite, NO<sub>2</sub><sup>-</sup>; phosphate, PO<sub>4</sub><sup>3-</sup>; and silicate, Si(OH)<sub>4</sub>—were collected at select locales concurrently with shallow-water and shipboard CTD casts.

## **A.2. Benthic Surveys and Collections, Monitoring Installations, and Microbial Sampling**

*(Jeff Anderson, Edmund Coccagna, Matthew Dunlap, Kerry Grimshaw, Jason Helyer, Steven Quistad, Russell Reardon, Cristi Richards, and Rodney Withall)*

CRED collected integrated information on the species composition (diversity), condition, abundance, and distribution of communities of corals, algae, and target macroinvertebrates and on benthic habitat complexity and substrates using 2 primary methodologies: Rapid Ecological Assessment (REA) surveys and towed-diver surveys. Performed at selected hard-bottom locations, REA benthic surveys include multiple methodologies that use two 25-m transect lines deployed at each REA site. Towed-diver surveys, which follow a depth contour of ~ 15 m and encompass various substrates, cover an area that is much broader than the area surveyed using fine-scale REA techniques. In addition, 3 types of monitoring installations, autonomous reef monitoring structure (ARMS), CAU, and EAR, serve as mechanisms to quantify marine invertebrates that are not easily identifiable during REA surveys, help to determine accretion rates of crustose coralline red algae and hard corals, or monitor the sounds of marine life and vessel traffic. Note that the sites where REA benthic surveys were conducted typically were different locations from the REA sites selected for fish surveys. REA sites for benthic surveys were selected for long-term monitoring of specific benthic communities over time, whereas REA sites for fish surveys were selected using a stratified random sampling design to provide representative coverage of 3 depth strata.

### **A.2.1. Benthic Composition**

Using a line-point-intercept (LPI) method at REA sites, hard corals, octocorals, macroalgae, crustose coralline red algae, turf algae, cyanobacteria, and macroinvertebrates were identified to the highest possible taxonomic resolution and recorded, along with sand cover, at 20-cm intervals along two 25-m transect lines set in a single file row (separated by 5 m). These surveys generate 125 points per transect (250 points per site) that can be used to generate percentage of cover of benthic organisms and sand at each REA site. Additionally, in concert with LPI surveys, still photographs were taken to record the benthos at intervals of 2 m along the same 2 transect lines, as well as at 5-, 15- and 25-m intercepts, with a high-resolution digital camera mounted on a pole. This work generates 30 photographs per site that will be later analyzed by staff at CRED, using the computer program Coral Point Count with Excel extensions (CPCe), to determine the benthic composition at higher taxonomic levels for each REA site (similar photographs of the benthos taken at REA sites surveyed by the fish team will also be analyzed).

Time permitting after LPI surveys were completed at each REA site, roving-diver surveys were conducted, covering a swath of 3–5 m on either side of the transect lines to record algal species richness.

If algal species encountered during LPI or roving-diver surveys were not identifiable in the field, an example was collected as a voucher specimen. These specimens were subsequently cataloged and critically analyzed to ensure positive species identification. Provisions were made to ensure appropriate preservation and curation of each algal specimen. These voucher specimens along with the benthic photographs form permanent historical records, the former of algal diversity and the latter of the composition of benthic communities at each REA site.

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 10 segments of 5 min each, 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 recorder 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. Benthic photos can be analyzed later for community structure information.

### **A.2.2. Community Structure and Disease**

At each REA site, the belt-transect method, with two 25-m transect lines as the focal point, was used to quantitatively assess generic richness, colony density, and size class of coral colonies. On each transect, five 2.5-m<sup>2</sup> segments were surveyed (0–2.5 m; 5.0–7.5



m; 10–12.5 m; 15–17.5 m; 20–22.5 m), whereby all coral colonies whose center fell within 0.5 m on either side of each transect line were identified to the highest possible taxonomic resolution and measured using 2 planar size metrics: maximum diameter and diameter perpendicular to the maximum diameter.

For each coral colony identified during belt-transect surveys, the extent of mortality, both recent and old, was estimated and signs of disease or compromised health were recorded, including type of lesion (bleaching, skeletal growth anomaly, white syndrome, tissue loss other than white syndrome, trematodiasis, necrosis, pigmentation responses, algal overgrowth, or other), extent (percentage of colony affected), and severity (mild, moderate, marked, severe, or acute). Levels of predation of corals were also recorded. In tandem with these coral disease surveys at each REA site, the belt-transect method also was used to quantify coralline-algal disease and syndromes, including coralline lethal orange disease, coralline white band syndrome, and coralline cyanobacterial disease.

### **A.2.3. Installations for Monitoring Marine Life**

CRED accomplishes long-term monitoring of benthic biodiversity, the growth rates of corals and algae, and the sounds of marine animals through the use of the following types of instruments that were retrieved or deployed during this cruise.

**Autonomous Reef Monitoring Structure (ARMS):** the ARMS is a long-term, collecting device designed to mimic the structural complexity of a coral reef habitat and attract colonizing invertebrates. The ARMS was developed specifically to monitor marine invertebrate cryptobiota that are not easily identifiable or accountable on the transect lines used for REA surveys. Each ARMS was composed of 10 grey, Type 1 PVC plates (23 × 23 cm) stacked in an alternating series of open and obstructed layers attached to a base plate (35 × 45 cm) that was affixed to a reef. ARMS were installed by pounding stainless steel rods by hand into bare substrate.

ARMS previously deployed during the Pacific RAMP cruise to Wake Atoll in 2009 were retrieved. Recovered ARMS were encapsulated with a mesh-lined lid, brought to the surface, and transported back to the ship for processing. Processing consisted of disassembling each ARMS unit, photo-documenting the sessile communities present on its plates, and preserving all captured organisms in ethanol for future molecular analyses. At a subsample of REA sites, new ARMS units were redeployed onto existing stainless steel rods, with the goal of recovering them during the next Pacific RAMP cruise scheduled for Wake Atoll in 2013.

**Calcification Acidification Unit (CAU):** deployed at several sites at each island, CAUs provide mechanisms to quantify accretion rates by crustose coralline red algae and scleractinian (hard) corals. Each CAU consists of 2 grey PVC plates (10 × 10 cm) separated by a 1-cm spacer. CAUs were installed on the benthos by pounding stainless steel rods by hand into bare substrate and then bolting plate assemblies to those rods. It has been demonstrated that PVC encourages growth of crustose coralline red algae and recruitment of corals, and the net weight gain of calcium carbonate (CaCO<sub>3</sub>) on the

surfaces of the CAUs can be an indicator of net calcification. The CAUs installed during this cruise will remain on the benthos for 2 years, enabling the recruitment and colonization of crustose coralline red algae and hard corals, upon which time they will be collected and analyzed. The data obtained via CAUs will enable a comparison of net calcification rates among islands and atolls and between archipelagos and form a baseline of accretion rates throughout the U.S. Pacific, allowing for future comparisons to determine possible consequences of increased ocean acidity and lowered aragonite saturation states.

**Ecosystem Acoustic Recorder (EAR):** the EAR is a passive acoustic device developed specifically for monitoring marine mammals, fishes, crustaceans, other sound-producing marine life, and human activity in marine habitats. The EAR is a digital, low-power system that records ambient sounds up to 30 kHz on a programmable schedule and can also respond to transient acoustic events that meet specific criteria, such as motorized vessels passing nearby or cetaceans. This type of subsurface instrument typically was deployed at depths of 5–25 m. Note: information about retrievals and deployments of EARs are provided along with information about STR installations in the island appendices, since those instruments were sometimes moored to the same anchor and EARs were typically installed by members of the oceanography team.

#### **A.2.4. Coral Core Collections**

The coring of massive coral colonies is aimed at studying coral growth and accretion rates to provide calcification and extension rate chronologies to hindcast the carbonate chemistry climate of coral reefs from hundreds of years past. To quantify the size and density of annual growth bands in coral skeletons, core samples were collected and preserved for analysis by nondestructive computerized axial tomography (CAT)-scan and image-analysis techniques to visualize growth bands that cannot otherwise be observed.

Dependant on ocean conditions, a maximum of 2 REA sites at each island was selected for coring. Collection of coral core samples targeted colonies of *Porites* spp., since their massive growth forms give them the greatest potential to provide long growth histories. At each REA site, a minimum of 2 and a maximum of 5 sample cores were collected in close proximity (3–5 m) to each other, using a small, handheld pneumatic drill operated from a scuba tank. The coring bit used was 35 cm long with an outer diameter of 3.8 cm and an inner diameter of 2.5 cm. Each core was collected from a single colony of sufficient size and health such that extracting a core 2.5 cm in diameter and 10–35 cm in length was judged not to be destructive or detrimental to the longevity of a colony. Through analyses of data from past CRED monitoring efforts, the abundance of coral colonies meeting coring criteria was established for each island, and cores were collected from areas where impact to coral populations was determined to be minimal. Upon completion of a coring, a cement plug was affixed to seal the hole, preventing invasion of bioeroding organisms. A cement plug provides a suitable surface over which surrounding coral tissue can grow. Coral core samples will be shipped to Woods Hole Oceanographic Institution in Woods Hole, Mass., for analyses using CAT-scan technology.

### **A.2.5. *Halimeda* Collections for Calcification Analysis**

Species of the green algal genus *Halimeda* are among the most important producers of calcified sediments in reef systems. As the acidity of our oceans increases, calcification rates and the ability of *Halimeda* algae to produce sediments may fall precipitously. To gain a baseline understanding of calcification levels in species of *Halimeda*, a joint project between CRED and the Scripps Institution of Oceanography, University of California San Diego, is sampling *Halimeda* populations across the Pacific to determine ambient levels of CaCO<sub>3</sub> among different species from different geographic areas. To accomplish this research, 10 individuals of 1 species of *Halimeda* were collected haphazardly by hand from established REA sites visited at Wake Atoll. Specimens were dried in an oven after collection and will be shipped to Scripps for analysis of percentage of calcification.

### **A.2.6. Microbial Communities and Water Chemistry**

Microbes are a fundamental aspect of all marine ecosystems. Trophic-level interactions within the marine microbial food web can have a big effect on global nutrient and carbon cycling. Within a reef system, the amount of energy from primary production remineralized by the microbial fraction determines the amount of energy available for the entire food web. Shifts in the abundance and composition of the microbial community in a reef system have also been linked to declines in coral health.

It is well known that bacteriophages (bacterial viruses) are the most abundant form of life in the ocean, ranging from  $1 \times 10^6$  virus-like particles (VLPs) per milliliter of seawater in the open ocean to  $1 \times 10^8$  VLPs per milliliter in more productive coastal waters. The number of microbial cells in seawater is typically  $1 \times 10^6$  cells per milliliter. Microbial and viral loading and the dominance of heterotrophic bacteria in reef water are linked to coral disease. The most direct method for assessing and monitoring changes in abundance of these microbiological components is by fluorescent microscopy using nucleic acid staining.

A direct parallel exists between microbial and viral loading, increasing human disturbance, and reef health. Microbial communities in more degraded coral reef systems support a high abundance of potential pathogens and heterotrophic microbes (a heterotrophic organism obtains food only from organic material, such as carbon and nitrogen, and is unable to use inorganic matter to form proteins and carbohydrates). In contrast, near-pristine reefs support microbial communities that are balanced between heterotrophs and autotrophs and contain very few potential pathogens (an autotrophic organism can synthesize food from inorganic material).

Spatial assessment of microbial and viral components with respect to levels of dissolved organic carbon (DOC), nutrients (NO<sub>2</sub><sup>-</sup>; NO<sub>3</sub><sup>-</sup>; PO<sub>4</sub><sup>3-</sup>; and ammonium, NH<sub>4</sub>), and particulate organic carbon (POC) within coral reef ecosystems may identify important predictors of coral reef ecosystem degradation. For example, in addition to microbial abundance, bacterial growth efficiency (BGE) may also play a role in reef system health.

BGE is greatly affected by DOC:Nitrogen ( $\text{NO}_x + \text{NH}_4$ ) ratios in the water column. Water column stoichiometry—carbon to nitrogen to phosphorous (C:N:P) ratios—directly affects microbial growth rates.

In summary, no long-term data on the dynamics of natural bacterial assemblages in reef systems (let alone other ecotypes) are currently available. Building a Pacific-wide microbial data set is an extremely important step towards greater understanding of the overall health of reef systems. The majority of reefs on the planet are affected, and analyses are confounded by the inability to attribute differences in reef system dynamics to variation in resource availability because of oceanography or human activity. The region monitored through Pacific RAMP includes reefs experiencing various combinations of human activity and resource availability. The hope is that new patterns in microbial data sets will emerge at regional or Pacific-wide scales and that this information can be used to understand the mechanisms underlying reef system decline.

**Collection of Microbial Water Samples:** At select REA sites, four 2-L samples of water were collected daily from < 1 m above the benthos using diver-deployable Niskin bottles. These water samples were returned to the ship, where they were processed for analyses of DOC, POC, particulate organic matter (POM), nutrients, microbial (bacteria and archaea) and viral abundance (fluorescent microscopy), fluorescence-activated cell sorting (FACS, heterotrophs vs. autotrophs), microbial and viral community composition (coarse analysis: 16s rRNA). At one REA site, ~ 80 L of water was collected from reef crevices and surfaces for metagenomic analysis of the microbial and viral community associated with reef benthos.

The following data items were collected daily at each REA site:

- DOC: 4 replicates
- POM: 4 replicates
- Nutrients: 2 replicates
- Microbial (bacteria and archaea) and viral abundance : 4 replicates (0.02- $\mu\text{m}$  filters, stained using SYBR Gold, Molecular Probes Inc., Eugene, Ore.)
- Microbial (bacteria and archaea) size structure : 4 replicates (0.2- $\mu\text{m}$  filters, stained using 4',6-Diamidino-2-phenylindole (DAPI))
- Microbial community composition (FACS, heterotrophs/autotrophs) : 5 replicates
- Microbial community composition (16s rRNA): 2 replicates (.22- $\mu\text{m}$  filters)

The following data items were collected once per island at REA sites:

- Microbial community composition (metagenome): 1 sample, (3–6 filters of 0.45  $\mu\text{m}$  each)
- Viral community composition (metagenome): 1 sample, (3–6 vials)

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

*Enumeration of microbes and viruses.* Replicate 5-mL and 500  $\mu$ L reef water samples were fixed using paraformaldehyde and filtered through 0.02- $\mu$ m filters. These filters were stained using the general nucleic acid stain SYBR Gold and mounted onto a microscope slide. Bacteria and VLPs were counted under UV light using Image Pro software.

*Microbial community size structure.* Replicate 5-mL samples of reef water were fixed with glutaraldehyde and filtered through 0.2- $\mu$ m 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 under UV light using Image Pro software. These slides can also be used to quantify the number of actively dividing microbial cells. Slide analysis will be performed at San Diego State University (SDSU). All filters will be stored at  $-20^{\circ}\text{C}$  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 provides complementary data for microbial abundance, microbial community structure, and levels of Chlorophyll *a*.

Five 1-mL samples of water from each REA site were pushed through a 20- $\mu$ m filter. This filtrate was dispensed into cryovials ( $5 \times 1$  mL) and fixed with glutaraldehyde. Vials were inverted to mix and incubated in the dark for 15 min. Glutaraldehyde-preserved samples were flash frozen in liquid nitrogen contained in a dry shipper to prevent damage to microbial cells. These samples were shipped upon return to Honolulu on dry ice to SDSU for flow cytometry analysis.

*Water Chemistry (DOC/POC):* 30 mL of seawater was filtered through pre-combusted glass fiber filters from each of the 4 Niskin bottles, and the filtrate was collected in precombusted glass bottles. Hydrochloric acid was added to each bottle to remove DIC, and the bottles were stored upright at  $4^{\circ}\text{C}$ . To assess POC, 500 mL of seawater was filtered through each glass fiber filter (4 replicates), and the filters were stored at  $-20^{\circ}\text{C}$ . Stable isotopes of carbon and nitrogen will also be analyzed from 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 via filtration followed by 454 sequencing. To remove large eukaryotic organisms, reef water was filtered through a 20- $\mu$ m pre-filter. This 20- $\mu$ m filtrate was subsequently passed through a 0.22- $\mu$ m Sterivex filter to trap microbes (2 filters,  $\sim 2.5$  L each). These

filters were stored at  $-20^{\circ}\text{C}$ . DNA isolation and metagenomic analysis will be completed at SDSU.

At one REA,  $\sim 80$  L of water was collected by filling four 20-L collapsible carboys with water from reef crevices or benthos using a manual bilge pump. Upon return to the ship, this sample was pre-filtered through 100- $\mu\text{m}$  mesh and concentrated using tangential flow filtration (TFF). TFF concentrates the bacteria and viruses in the water, bringing the initial  $\sim 80$  L of water to a final volume of  $\sim 500$  mL. This concentrate was then filtered through 0.45- $\mu\text{m}$  filters to capture microbes (bacteria and archaea). These filters were then frozen. 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 made it through the 0.45- $\mu\text{m}$  filter, and this sample was stored at  $4^{\circ}\text{C}$ . 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 Benthos-associated Microbial DNA:** This section describes samples, or benthic grabs, collected if time permitted. The following data items were collected at REA sites when time permitted:

- Rubble and sediment: several replicate bags
- Algae: several replicate bags

In addition to monitoring changes in microbial communities associated with the water column, this work is investigating whether or not community shifts in the microbes associated with the benthos are a useful indicator of reef health. When time permitted, several “fist-fulls” of rubble and sediment and several pieces of the most dominant type of algae were collected in Ziploc bags. These samples were frozen at  $-20^{\circ}\text{C}$  and will stay on the ship until it returns to Honolulu. The 16s bacterial rRNA genes associated with these samples will be sequenced to characterize the microbial communities associated with the benthos (rubble and algae).

### **A.3. Surveys of Reef Fishes**

*(Paula Ayotte, Brian Hauk, Kevin Lino, Kaylyn McCoy, Kara Osada-D’Avella, and Ben Richards)*

Four divers conducted REA fish surveys using the stationary-point-count (SPC) method at preselected REA sites. Two separate teams performed these surveys. Each team consisted of 2 divers and conducted either 1 or 2 SPC surveys 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. 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. Every 2 m along the transect line, still photographs were taken of the benthos at a distance of 1 m from the right side of the line. If only one replicate survey was completed at a REA site (because of insufficient air pressure or bottom time), benthic photographs were taken at each meter mark. Like the photographs taken along transect lines during surveys at REA benthic sites, these images will be analyzed later.

If bottom time and air permitted, the 30-m transect line was moved to another location 5–10 m away at the same depth stratum, and this procedure was repeated.

In addition to site-specific REA surveys, broad-scale towed-diver surveys were used to characterize the fish communities of shallow-water habitats around each island. A pair of divers, by means similar to a manta-tow technique, was towed 60 m behind a small boat, a 6-m survey launch from SAFE Boats International, with one diver quantifying fish populations and the other quantifying the benthos. Each towed-diver survey lasted 50 min, broken into 10 segments of 5-min each, 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 GPS unit on the boat. A custom algorithm was used to calculate the track of the divers based on the track, 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 recorder set 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 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<sup>-2</sup>). 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 or grounding of a vessel.



## APPENDIX B: WAKE ATOLL

Wake Atoll is located at 19°36' N, 166°30' E in the North 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.”

### B.1. Oceanography and Water Quality

Oceanographic operations during the cruise HA-11-01, Leg I, at Wake Atoll entailed numerous retrievals and deployments of oceanographic moored instruments, installation of calcification acidification units (CAUs), nearshore water sampling and conductivity, temperature, and depth (CTD) casts at Rapid Ecological Assessment (REA) sites, shipboard water sampling and CTD casts offshore to a depth of 500 m, and acoustic Doppler current profiler (ADCP) transect lines.

Seven subsurface temperature recorders (STRs) were retrieved and deployed for long-term monitoring (Fig. B.1.1 and Table B.1.1). Two acoustic Doppler profilers (ADPs), 6 SBE 37 temperature and salinity sensors, and 3 STRs were recovered and not replaced; these instruments were part of a short-term investigative research study (see the next paragraph for more information about this study). Four STRs were deployed where SBE 37 sensors were previously located, establishing new long-term monitoring sites. One sea-surface temperature (SST) buoy and SST anchor were recovered and replaced from the lagoon, while an ecological acoustic recorder (EAR) and colocated STR were recovered only (Table B.1.1). One HARP was recovered and replaced. For information about CAU deployments completed at Wake Atoll, see Section B.2: “Benthic Environment.”

Temporary installments of oceanographic moorings were deployed during the previous Pacific RAMP cruise (HA-09-01) to Wake Atoll in 2009 as part of a site-specific study looking at biophysical interactions on coral reefs. These mooring deployments consisted of 2 cross-shelf transects on the northwest and southeast sides of Wake Atoll. Each transect consists of an ADP with directional wave capabilities, an SBE 37 sensor at a depth of 20 m, and STRs at depths of 15 and 10 m. An additional 4 SBE 37 sensors were deployed at a depth of 20 m at various locations around this atoll. All moorings were recovered except 1 STR, which was deemed lost. This study is a collaborative project between CRED, the University of Hawai`i, and the Scripps Institution of Oceanography with the purpose of understanding the impacts of physical oceanographic forcing on benthic coral reef community structure.

At nearshore locations around Wake Atoll, 5 shallow-water CTD casts were performed (Fig. B.1.1) at select REA sites where CAUs were installed. In concert with each CTD cast, 2 water samples were taken to measure the following parameters: dissolved inorganic carbon (DIC), total alkalinity (TA), salinity, chlorophyll-*a* (Chl-*a*), and nutrient concentrations. Accounting for losses and microbiological nutrients taken alone, a total of

10 DIC and TA, 10 salinity, 10 Chl-*a*, and 12 nutrient water samples were collected, 1 from the surface and 1 near the reef at each REA site where CAUs were installed. In addition to the standard discrete water sampling, a remote auto sampler (RAS) was deployed at 1 REA site (WAK-08) to collect hourly water samples over a 48-h period for DIC and TA measurements (Fig. B.1.1). A CTD sensor and ADP attached to that RAS collected measurements of salinity, temperature, and current at 1-min intervals during the same 48-h period.

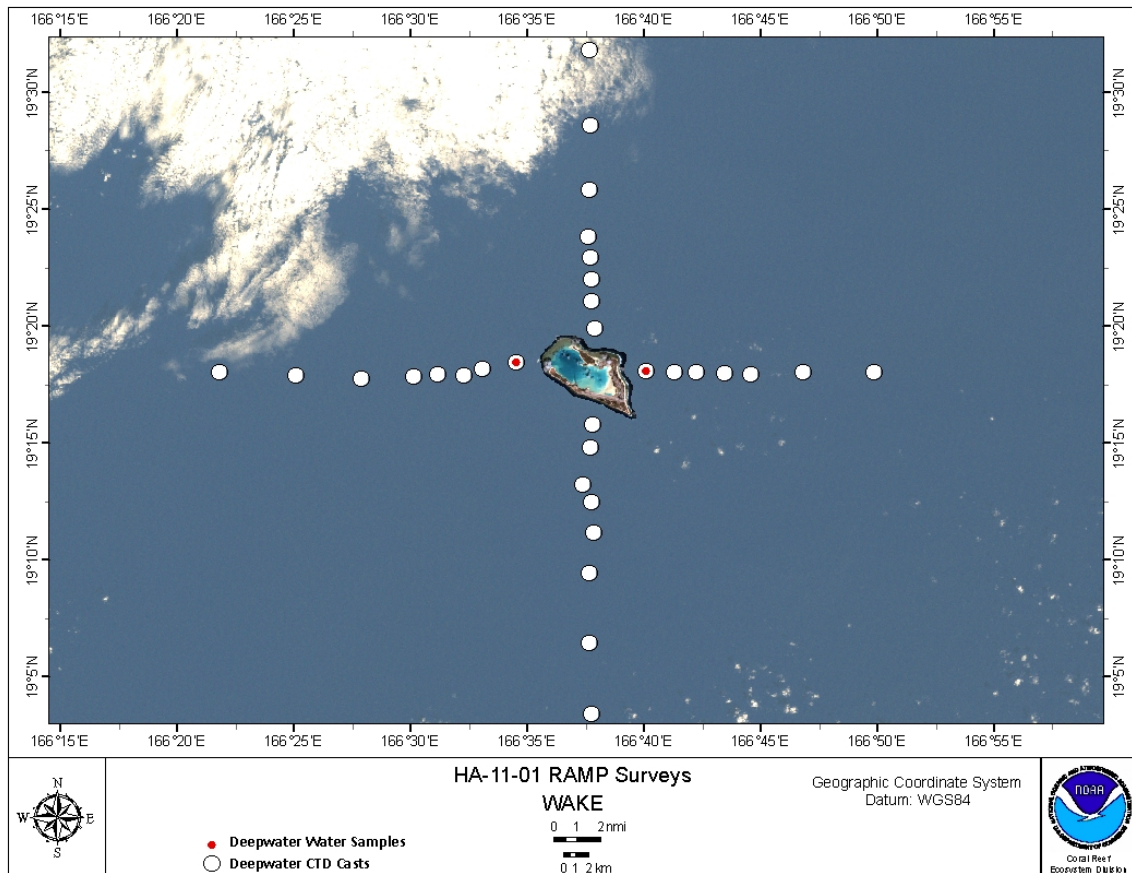
From the NOAA Ship *Hi'ialakai*, ~ 100 km of ADCP transect lines were run in the 4 cardinal directions away from this island during night operations. On the reciprocal course, shipboard CTD casts were conducted to a depth of 500 m per transect line every 2–4 km for a total of 31 deepwater CTD casts. Water samples were collected concurrently with 2 select shipboard CTD casts at 5 depths between the surface and 200 m, depending on the depth of mixed layer as determined by the CTD downcast (Fig. B.1.2). Near Wake Atoll, 10 Chl-*a* and 10 nutrient shipboard water samples were collected.



**Figure B.1.1.**--Mooring sites at Wake Atoll where oceanographic instruments were retrieved or deployed and locations of nearshore CTD casts and water sampling conducted during cruise HA-11-01, Leg I (IKONOS Carterra Geo Data, 2003; Landsat satellite imagery data used in this map figure are available from the U.S. Geological Survey).

**Table B.1.1.--**Geographic coordinates and sensor depths of the moored oceanographic instruments, EARs, and anchors that were retrieved or deployed at Wake Atoll during cruise HA-11-01, Leg I.

Mooring Site	Date	Instrument Type	Latitude	Longitude	Depth (m)	Retrieved	Deployed
WAK-006	23-Mar	ADP	19.31626844	166.59832360	10.7	–	1
WAK-006	23-Mar	CTD	19.31626844	166.59832360	10.7	–	1
WAK-006	23-Mar	RAS	19.31626844	166.59832360	10.7	–	1
WAK-006	23-Mar	STR	19.31625662	166.59836940	11	1	1
WAK-010	23-Mar	ADP	19.32585717	166.60981160	18.3	1	–
WAK-010	23-Mar	SBE37	19.32585717	166.60981160	18.3	1	–
WAK-011	23-Mar	STR	19.32568652	166.60970230	13.4	1	–
WAK-004	24-Mar	STR	19.30620840	166.65099580	14.6	1	1
WAK-005	24-Mar	STR	19.30626984	166.65150870	25.9	1	1
WAK-007	24-Mar	STR	19.31631446	166.59797430	24.4	1	1
WAK-013	24-Mar	ADP	19.27674794	166.65720410	20.4	1	–
WAK-013	24-Mar	SBE37	19.27674794	166.65720410	20.4	1	–
WAK-013	24-Mar	STR	19.27674794	166.65720410	20.4	–	1
WAK-014	24-Mar	STR	19.27672153	166.65700120	14.3	1	–
WAK-015	24-Mar	STR	19.27668164	166.65678880	10.7	1	–
WAK-016	24-Mar	SBE37	19.30632340	166.65151490	20.7	1	–
WAK-001	25-Mar	STR	19.28032634	166.62867150	12.2	1	1
WAK-002	25-Mar	SST	19.30735923	166.62214360	0.3	1	1
WAK-003	25-Mar	STR	19.30867586	166.60449770	1.8	1	1
WAK-008	25-Mar	EAR	19.30093962	166.63801250	0.9	1	–
WAK-008	25-Mar	STR	19.30093962	166.63801250	0.9	1	–
WAK-009	25-Mar	Anchor	19.30735923	166.62214360	3.2	1	1
WAK-009	25-Mar	STR	19.30735923	166.62214360	3.2	1	1
WAK-018	25-Mar	SBE37	19.30295471	166.59486380	18.3	1	–
WAK-018	25-Mar	STR	19.30295471	166.59486380	18.3	–	1
WAK-019	25-Mar	SBE37	19.28021067	166.62839370	19.8	1	–
WAK-019	25-Mar	STR	19.28021067	166.62839370	19.8	–	1
WAK-006	26-Mar	ADP	19.31625964	166.59832820	10.7	1	–
WAK-006	26-Mar	CTD	19.31625964	166.59832820	10.1	1	–
WAK-006	26-Mar	RAS	19.31625964	166.59832820	10.7	1	–
WAK-017	26-Mar	SBE37	19.31624992	166.62714990	20.1	1	–
WAK-017	26-Mar	STR	19.31623994	166.62724070	20.7	–	1



**Figure B.1.2.**--Locations of shipboard CTD casts and water sampling performed at Wake Atoll during cruise HA-11-01, Leg I (IKONOS Carterra Geo Data, 2003; Landsat satellite imagery data used in this map figure are available from the U.S. Geological Survey).

## B.2. Benthic Environment

Belt-transect and line-point-intercept (LPI) surveys were conducted and photographs were taken along transect lines at 11 REA sites at Wake Atoll to assess benthic composition, coral and algal community structure, and coral and algal disease (Fig. B.2.1 and Table B.2.1). At the end of LPI surveys, roving-diver algal surveys were conducted at all REA sites.

Various samples were collected at 8 REA sites (Table B.2.1): 6 algal voucher specimens at 4 REA sites for taxonomic identification, 30 individuals of the algal genus *Halimeda* at 3 REA sites for calcification analysis, 2 coral cores (20–35 cm in length) from *Porites lobata* coral heads at 2 REA sites for calcification research, and 20 water samples for microbial analyses at 4 REA sites with 4 water samples of 2 L each at each site and 4 water samples of 20 L each at 1 site, WAK-06. Additional microbial work included benthic grabs of coral rubble, sediment, and individuals of the algal genus *Halimeda* at 1 REA site. For more information about collections made at REA sites, see Table C.1 in Appendix C: “Biological Collections.”



Nine autonomous reef monitoring structures (ARMS) were recovered: 3 ARMS each from WAK-06, WAK-09, and WAK-10 (Table B.2.1). Nine ARMS were deployed with 3 ARMS each at WAK-06, WAK-08, and WAK-09. At each of 5 select REA sites, an array of 5 CAUs was deployed for a total of 25 CAUs installed at Wake Atoll (Table B.2.1). For information about a retrieval of an EAR, see Section B.2: “Oceanography and Water Quality.”

In total, the benthic team conducted 75 individual dives at REA sites around Wake Atoll.



**Figure B.2.1.**--Locations of REA benthic sites surveyed at Wake Atoll during cruise HA-11-01, Leg I (IKONOS Carterra Geo Data, 2003; Landsat satellite imagery data used in this map figure are available from the U.S. Geological Survey).

**Table B.2.1.**--Summary of REA benthic surveys, CAU installations, and ARMS retrievals (Ret.) and deployments (Dep.) performed as well as algal specimens and microbial water and benthic samples collected at Wake Atoll during cruise HA-11-01, Leg I. Indication that an LPI survey was completed also means that photographs were taken along transect lines and roving-diver algal surveys were conducted. Counts of algal samples include both algal voucher specimens and *Halimeda* samples for calcification analysis.

REA Site	Date	Latitude	Longitude	REA Surveys		Installations and Collections			
				LPI	Corals	CAUs	ARMS Ret/Dep	Algae	Microbial Samples
WAK-08	23-Mar	19.3162710	166.5983410	×	×	5	0/3	0	0
WAK-13	23-Mar	19.3154022	166.6433940	×	×	5	0/0	0	4
WAK-03	24-Mar	19.2976289	166.5982870	×	×	0	0/0	10	0
WAK-05	24-Mar	19.3028400	166.5948600	×	×	0	0/0	11	0
WAK-06	24-Mar	19.2917761	166.6072900	×	×	5	3/3	0	8
WAK-01	25-Mar	19.2806617	166.6278300	×	×	5	0/0	1	0
WAK-04	25-Mar	19.3078616	166.5938220	×	×	0	0/0	1	4
WAK-09	25-Mar	19.2706753	166.6516390	×	×	0	3/3	0	0
WAK-02	26-Mar	19.2878044	166.6128680	×	×	0	0/0	0	0
WAK-07	26-Mar	19.3204600	166.602140	×	×	0	0/0	13	27
WAK-09	26-Mar	19.2706787	166.6515780	–	–	5	0/0	0	0
WAK-10	26-Mar	19.3101300	166.6038300	–	–	0	3/0	0	0
WAK-12	26-Mar	19.3254000	166.6118600	×	×	0	0/0	0	0

CRED also completed 18 towed-diver surveys at Wake Island, covering a total length of 36.1 km (an area of 36.1 ha) on the ocean floor (Fig. B.2.2). The mean survey length was 2.0 km with a range of 1.4–2.5 km. The mean survey depth was 14.5 m with a range of 12.5–17.2 m. The mean temperature from data recorded during these surveys was 26.1°C with a range of 25.4°C–26.6°C.

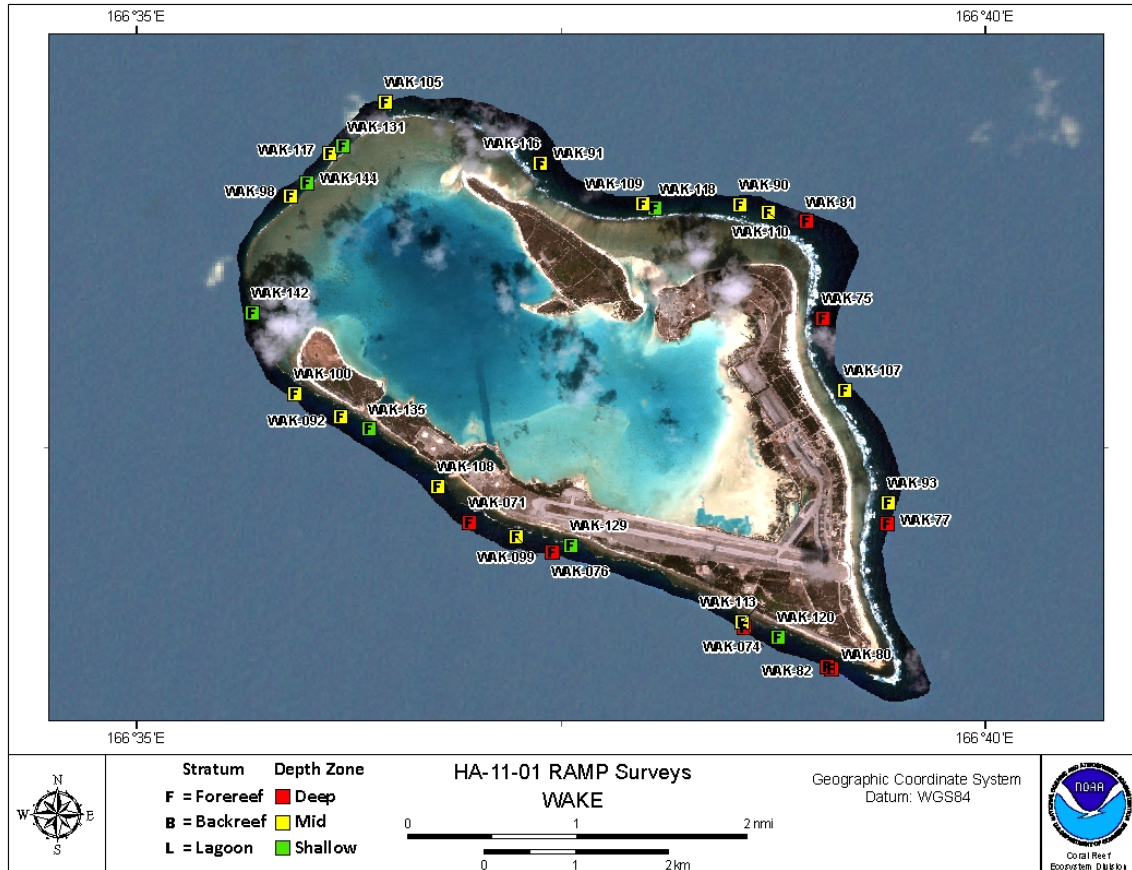


**Figure B.2.2.**--Track locations of towed-diver surveys conducted around Wake Island during cruise HA-11-01, Leg I (IKONOS Carterra Geo Data, 2003; Landsat satellite imagery data used in this map figure are available from the U.S. Geological Survey).

### B.3. Reef Fish Community

REA fish survey sites were chosen using a stratified random design. Stationary-point-count surveys were conducted at 30 REA sites around Wake Atoll in the deep, moderate, and shallow foreereef strata (Table B.3.1 and Fig.B.3.1). No fishes were collected during these surveys.

In addition, CRED completed a total of 18 towed-diver surveys at Wake Atoll, as described previously in Section B.2 of this appendix.



**Figure B.3.1.**--Locations of REA fish sites surveyed at Wake Atoll during cruise HA-11-01, Leg I. All of these REA sites were selected using a stratified random design (IKONOS Carterra Geo Data, 2003; Landsat satellite imagery data used in this map figure are available from the U.S. Geological Survey).



**Table B.3.1.--**Summary of sites where REA fish surveys were conducted at Wake Atoll during cruise HA-11-01, Leg I.

REA Site	Date	Depth Zone	Stratum	Depth (m)	Latitude	Longitude
WAK-91	23-Mar	Moderate	Forereef	15	19.31941417	166.62303510
WAK-98	23-Mar	Moderate	Forereef	12	19.31633843	166.59841160
WAK-100	23-Mar	Moderate	Forereef	17	19.29691563	166.59879470
WAK-117	23-Mar	Moderate	Forereef	14	19.32048270	166.60225070
WAK-142	23-Mar	Shallow	Forereef	4	19.30485522	166.59461530
WAK-144	23-Mar	Shallow	Forereef	5	19.31762522	166.59998840
WAK-71	24-Mar	Deep	Forereef	24	19.28428897	166.61594080
WAK-74	24-Mar	Deep	Forereef	23	19.27399591	166.64301340
WAK-76	24-Mar	Deep	Forereef	24	19.28137056	166.62411200
WAK-92	24-Mar	Moderate	Forereef	14	19.29461472	166.60334140
WAK-99	24-Mar	Moderate	Forereef	16	19.28282188	166.62058890
WAK-129	24-Mar	Shallow	Forereef	5	19.28197288	166.62589880
WAK-135	24-Mar	Shallow	Forereef	4	19.29341250	166.60607360
WAK-75	25-Mar	Deep	Forereef	24	19.30428819	166.65066340
WAK-77	25-Mar	Deep	Forereef	23.5	19.28411697	166.65699780
WAK-80	25-Mar	Deep	Forereef	25.1	19.26983622	166.65157430
WAK-93	25-Mar	Moderate	Forereef	16	19.28616584	166.65709200
WAK-107	25-Mar	Moderate	Forereef	14.4	19.29726105	166.65284190
WAK-113	25-Mar	Moderate	Forereef	14.3	19.27449354	166.64281280
WAK-120	25-Mar	Shallow	Forereef	5	19.27304297	166.64632160
WAK-81	26-Mar	Deep	Forereef	24.2	19.31377717	166.64908600
WAK-90	26-Mar	Moderate	Forereef	11.5	19.31542941	166.64250260
WAK-105	26-Mar	Moderate	Forereef	14	19.32548669	166.60769920
WAK-109	26-Mar	Moderate	Forereef	14	19.31553679	166.63304820
WAK-110	26-Mar	Moderate	Forereef	13	19.31466356	166.64535430
WAK-116	26-Mar	Moderate	Forereef	15	19.31953311	166.62289070
WAK-118	26-Mar	Shallow	Forereef	5.5	19.31508232	166.63414740
WAK-131	26-Mar	Shallow	Forereef	5.2	19.32123195	166.60350440
WAK-82	27-Mar	Deep	Forereef	25	19.27007628	166.65111180
WAK-108	27-Mar	Moderate	Forereef	15	19.28780509	166.61283980

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## APPENDIX C: BIOLOGICAL COLLECTIONS

Biological samples were collected at Wake Atoll for multiple research purposes. A complete listing of these collections is presented here in Table C.1

**Table C.1.**--Biological samples collected at Wake Atoll during cruise HA-11-01, Leg I.

REA Site	Date	Latitude	Longitude	Specimen Collected	Number of Samples	Depth (m)
<b>Algal Collections: Calcification Analysis</b>						
WAK-03	24-Mar	19.2976300	166.5982800	<i>Halimeda</i> sp.	10	13.7
WAK-05	24-Mar	19.3028400	166.5948600	<i>Halimeda</i> sp.	10	13.7
WAK-07	26-Mar	19.3204600	166.6021400	<i>Halimeda</i> sp.	10	13.7
<b>Algal Collections: Voucher Specimens</b>						
WAK-05	24-Mar	19.3028400	166.5948600	Red Alga	1	13.7
WAK-01	25-Mar	19.2806617	166.6278300	Red Alga	1	13.7
WAK-04	25-Mar	19.3078616	166.5938220	Red Alga	1	14.6
WAK-07	26-Mar	19.3204600	166.6021400	Red Algae	3	13.7
<b>Coral Collections: Cores</b>						
WAK-08	27-Mar	19.3164504	166.5985260	<i>Porites lobata</i>	1	15.5
WAK-05	27-Mar	19.3030054	166.5949710	<i>Porites lobata</i>	1	14.9
<b>Microbial Collections: Water Samples and Benthic Grabs</b>						
WAK-13	23-Mar	19.3154022	166.6433940	2 L	4	13.7
WAK-06	24-Mar	19.2917761	166.6072900	20L	4	13.7
WAK-06	24-Mar	19.2917761	166.6072900	2 L	4	13.7
WAK-04	25-Mar	19.3078616	166.5938220	2 L	4	13.7
WAK-07	26-Mar	19.3204600	166.6021400	2 L	4	10.7
WAK-07	26-Mar	19.3204600	166.6021400	Coral Rubble	10	10.7
WAK-07	26-Mar	19.3204600	166.6021400	<i>Halimeda</i> sp.	10	10.7
WAK-07	26-Mar	19.3204600	166.6021400	Sediment	3	10.7