

**Papahānaumokuākea Marine National Monument**  
RESEARCH Permit Application

**NOTE:** *This Permit Application (and associated Instructions) are to propose activities to be conducted in the Papahānaumokuākea Marine National Monument. The Co-Trustees are required to determine that issuing the requested permit is compatible with the findings of Presidential Proclamation 8031. Within this Application, provide all information that you believe will assist the Co-Trustees in determining how your proposed activities are compatible with the conservation and management of the natural, historic, and cultural resources of the Papahānaumokuākea Marine National Monument (Monument).*

**ADDITIONAL IMPORTANT INFORMATION:**

- Any or all of the information within this application may be posted to the Monument website informing the public on projects proposed to occur in the Monument.
- In addition to the permit application, the Applicant must either download the Monument Compliance Information Sheet from the Monument website OR request a hard copy from the Monument Permit Coordinator (contact information below). The Monument Compliance Information Sheet must be submitted to the Monument Permit Coordinator after initial application consultation.
- Issuance of a Monument permit is dependent upon the completion and review of the application and Compliance Information Sheet.

**INCOMPLETE APPLICATIONS WILL NOT BE CONSIDERED**

Send Permit Applications to:

Papahānaumokuākea Marine National Monument Permit Coordinator  
6600 Kalaniana'ole Hwy. # 300  
Honolulu, HI 96825

[nwhipermit@noaa.gov](mailto:nwhipermit@noaa.gov)

PHONE: (808) 397-2660      FAX: (808) 397-2662

**SUBMITTAL VIA ELECTRONIC MAIL IS PREFERRED BUT NOT REQUIRED. FOR ADDITIONAL SUBMITTAL INSTRUCTIONS, SEE THE LAST PAGE.**

## **Papahānaumokuākea Marine National Monument Permit Application Cover Sheet**

This Permit Application Cover Sheet is intended to provide summary information and status to the public on permit applications for activities proposed to be conducted in the Papahānaumokuākea Marine National Monument. While a permit application has been received, it has not been fully reviewed nor approved by the Monument Management Board to date. The Monument permit process also ensures that all environmental reviews are conducted prior to the issuance of a Monument permit.

### **Summary Information**

**Applicant Name:** Greta Smith Aeby

**Affiliation:** HIMB

**Permit Category:** Research

**Proposed Activity Dates:** May 1 - Sept 30, 2010

**Proposed Method of Entry (Vessel/Plane):** NOAA research vessel Hiialakai

**Proposed Locations:** shallow water reefs throughout the Monument

**Estimated number of individuals (including Applicant) to be covered under this permit:**

2

**Estimated number of days in the Monument:** 21-28 days

**Description of proposed activities:** (complete these sentences):

a.) The proposed activity would...

Determine the incidence of coral disease at several sites within the Monument and test a method for managing damage from Acropora growth anomalies. Fish (surgeonfish and butterflyfish) with skin cancer would be surveyed to determine distribution and prevalence of the disease in fish populations. Determine the effect of skin cancer on the body condition of fish. Baseline studies on the parasites of zooplankton will be initiated.

b.) To accomplish this activity we would ....

Survey reefs for coral disease, mark and photograph individual colonies exhibiting signs of disease, repair permanent sites, surgically remove growth anomalies off of table corals to determine efficacy of this method for managing disease. Surgeonfish and butterflyfish populations will be surveyed. Surgeonfish with skin cancer would be collected to examine the effect of the disease on the body condition of the fish. Plankton tows and light traps will be used to collect zooplankton at the different islands to be screened for parasite infections.

c.) This activity would help the Monument by ...

giving them information as to the health status of their reefs, ability to predict damage from coral disease through time, and a potential method to control *Acropora* growth anomalies. Studies of fish disease will give them information on how widespread cancer is in fish population within the Monument.

**Other information or background:** Global climate change and human activities are placing coral reef ecosystems at risk. Coral reefs worldwide are now declining at an alarming rate. Mass bleaching events have increased dramatically since the 1980's and have usually been linked to El Niño or global warming-related increases in annual sea surface temperature (Brown 1997, Barber et al. 2001). The El Niño Southern Oscillation (ENSO) conditions during 1997 to 1998 resulted in worldwide bleaching from the Western Atlantic to the Great Barrier Reef. ENSO events have increased in frequency and duration in the past two decades (Barber et al. 2001, Walker 2001) and it has been predicted that the frequency and severity of coral bleaching will also continue to rise (Hoegh-Guldberg 1999).

Disease in coral reef ecosystems has received great attention, particularly in the western Atlantic where coral disease has been incriminated in the marked degradation of reef habitats. (Santavy and Peters 1997, Green and Bruckner 2000). Coral disease is reported to be responsible for the dramatic decline of *Acroporids*, one of the major frame-building corals in the Florida Keys, changing the structure and function of the coral reef ecosystem (Aronson & Precht 2001). Despite the major impact disease can have on reef systems, the etiology of most coral diseases remains unclear (Santavy and Peters 1997, Richardson 1998). The causative agents, mechanism of pathogenesis and link to environmental or anthropogenic stress are still largely unknown (Richardson 1998, Green & Bruckner 2000).

The reefs of the Northwestern Hawaiian Islands (NWHI) are considered to be relatively healthy but they are not immune to the conditions that have led to the decline of other reef systems. In September 2002 the first mass-bleaching event was recorded on the reefs of the NWHI with a second bleaching event occurring in 2004. In the three northwestern most atolls of the Archipelago (Pearl & Hermes, Midway and Kure) over half of all sites had significant bleaching (Aeby et al. 2003, Kenyon et al., 2005). Ten coral disease states have now been described from the NWHI (Aeby 2006) and we have established permanent sites which allow us to determine both temporal and spatial changes in diseases through time and the ultimate affect of disease on the health of the ecosystem. We will measure changes in disease levels through time, rates of tissue loss from different diseases, patterns of disease transmission among colonies, rate of spread of disease and evaluate changes in coral cover and coral species composition. In addition, two diseases of concern have been identified, *Acropora* white syndrome and *Acropora* growth anomalies which we are targeting for focused studies.

*Acropora* white syndrome (AWS) is a disease which causes acute tissue loss in *acroporids* and has been reported from across the Indo-Pacific. *Acropora* white syndrome appeared on one reef in the northwestern Hawaiian Islands (NWHI) in 2003 (Aeby 2006) and has since spread. Our prior studies in 2005 and 2006 found this disease to be highly virulent having killed over 19 large table *acroporids* with numerous other colonies suffering massive tissue loss from the disease. The disease occurs predominantly at French Frigate Shoals (FFS) within the NWHI, which is the center of abundance and diversity of *acroporids* in Hawaii. We plan to continue to follow the dynamics of this disease by re-surveying permanent sites to measure coral mortality and disease spread.

Disease can affect coral communities directly through mortality of colonies (partial or whole) resulting in reduced coral cover (such as we found for AWS) or indirectly through sub-lethal events such as reduced growth, resilience or reproduction. From our 2006 study we discovered that *Acropora cytherea* with growth anomalies suffer a significant reduction in reproductive output. We would now like to determine whether this disease also affects the growth of colonies and whether removal of growth anomalies could be an effective management tool. During our prior studies we documented the occurrence of "dead zones" within "tumor city" at one of our permanent sites. This suggest that this disease is slowing killing corals through time. By tagging individual affected colonies for growth studies we will also be able to determine the lethality of this disease through time.

Diseases in marine ecosystems are not only limited to corals. Fibropapillomatosis of green turtles has been known in Hawaii since the 1950s (Balaz 1991). More recently, high levels of infections with bacteria and protozoa have been seen in taape (*Lutjanus kasmira*) (Work et al. 2003). Taape were introduced into Hawaii in the 1950s (Randall 1987) and have spread all the way to Midway Atoll. Taape are closely associated with certain native fish such as goatfish (*Mulloidichthys* sp.) (Friedlander et al. 2002) and goatfish from the main Hawaiian Islands have been found to be infected with some of the same diseases as taape (Work et al. unpub. data). Given that taape were introduced into Hawaii, there is the concern that the recently documented diseases may have been introduced with them from the Marquesas. Taape are infected with a parasitic gut nematode that is thought to have been brought into the Hawaiian ecosystem with the introduction of the fish. This nematode infection has also been found in co-occurring native goatfish species. Taape were originally introduced into Oahu and have recruited out to other islands and up into the NWHI. The question now arises as to whether disease transmission has occurred from the main HI out to the NWHI.

From our 2006 study we found that taape from FFS had the nematode infection yet this disease was not found in fish from Midway. It appears that there is a lag in the time required for taape to establish in the NWHI as compared to the establishment of fish disease. The spread of both taape and its diseases up into the NWHI may be reflective of real time ecological linkages between islands within the Hawaiian archipelago. We have a rough timeline of the spread of taape from Oahu out to Midway and could correlate that with the eventual emergence of this disease at Midway. From studies in 2006, we also found that species of native goatfishes from FFS also have the nematode infection. We would like to also sample goatfishes from the other islands we are visiting to determine whether the pattern of disease is similar to that found in taape.

Based upon studies of similar nematodes, we hypothesized that the first intermediate host of the potentially introduced parasitic red nematode is a planktonic copepod which could be the mechanism of disease dispersal throughout the Hawaiian Islands. Preliminary work at our lab in Honolulu has potentially identified the copepod hosts species as *Lobidocera madurae* and *Undinula vulgaris*. During this study we also found that the zooplankton in Kaneohe Bay contained a variety of different larval parasites. Parsites transmitted through the foodchain, such as these within the copepods, are useful as potential indicators of ecosystem health. Parasites can only remain in host populations if all hosts within the life cycle are present and in sufficient numbers to maintain the disease. Hence, healthy ecosystems may have higher levels of certain types of parasites as compared to more degraded regions where host abundance is reduced. We would now to compare the parasite levels of the zooplankton within the NWHI with our finding

within the MHI and explore whether there is evidence that copepods infected with the larval stage of the red nematode may be the mechanism of disease spread within the Hawaiian Archipelago.

From our 2005 and 2006 studies we found that the surgeonfish, *Ctenochaetus strigosus*, (kole) with a pigment discoloration had pathology consistent with cancerous lesions. Further survey work within the MHI found diseased kole on Oahu, Maui, Kauai and Molokai. We also observed several other species of surgeonfish with similar patterns of discoloration. Using the same survey method used in the MHI we would now like to compare distribution and prevalence of diseased fish in the NWHI.

It is important for management agencies to have a through understanding of the vulnerability of these reefs to disease and the first steps in managing disease are developing an understanding of the causes of disease and assessing its geographic extent. Mangement of disease in wildlife populations usually involves either reducing or removing the source of infection or reducing the spread of the disease. However, before appropriate management plans can be made the epizootiology of diseases must be understood. Our studies, past and proposed, are suppling critical information into disease dynamics in both coral and fish within the NWHI.

## **Section A - Applicant Information**

### **1. Applicant**

Name (last, first, middle initial): Aeby, Greta S.

Title: assistant researcher

#### **1a. Intended field Principal Investigator (See instructions for more information):**

Dr. Greta Aeby

#### **2. Mailing address (street/P.O. box, city, state, country, zip):**

[REDACTED]

[REDACTED]

[REDACTED]

[REDACTED]

For students, major professor's name, telephone and email address:

#### **3. Affiliation (institution/agency/organization directly related to the proposed project):**

Hawaii Institute of Marine Biology

#### **4. Additional persons to be covered by permit. List all personnel roles and names (if known at time of application) here (e.g. John Doe, Research Diver; Jane Doe, Field Technician):**

Dr. Fenny Cox: co-investigator

Dr. Frank Stanton: co-investigator



**Section B: Project Information**

**5a. Project location(s):**

<input type="checkbox"/> Nihoa Island	<input type="checkbox"/> Land-based	<input type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input type="checkbox"/> Necker Island (Mokumanamana)	<input type="checkbox"/> Land-based	<input type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> French Frigate Shoals	<input type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> Gardner Pinnacles	<input type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> Maro Reef			
<input checked="" type="checkbox"/> Laysan Island	<input type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> Lisianski Island, Neva Shoal	<input type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> Pearl and Hermes Atoll	<input type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> Midway Atoll	<input type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input checked="" type="checkbox"/> Kure Atoll	<input type="checkbox"/> Land-based	<input checked="" type="checkbox"/> Shallow water	<input type="checkbox"/> Deep water
<input type="checkbox"/> Other			

**Ocean Based**

NOTE: There is a fee schedule for people visiting Midway Atoll National Wildlife Refuge via vessel and aircraft.

Location Description:

shallow reefs throughout the Monument

**5b. Check all applicable regulated activities proposed to be conducted in the Monument:**

- Removing, moving, taking, harvesting, possessing, injuring, disturbing, or damaging any living or nonliving Monument resource
- Drilling into, dredging, or otherwise altering the submerged lands other than by anchoring a vessel; or constructing, placing, or abandoning any structure, material, or other matter on the submerged lands
- Anchoring a vessel
- Deserting a vessel aground, at anchor, or adrift
- Discharging or depositing any material or matter into the Monument
- Touching coral, living or dead
- Possessing fishing gear except when stowed and not available for immediate use during passage without interruption through the Monument
- Attracting any living Monument resource
- Sustenance fishing (Federal waters only, outside of Special Preservation Areas, Ecological Reserves and Special Management Areas)
- Subsistence fishing (State waters only)
- Swimming, snorkeling, or closed or open circuit SCUBA diving within any Special Preservation Area or Midway Atoll Special Management Area

**6 Purpose/Need/Scope *State purpose of proposed activities:***

1. To re-survey permanent sites established in 2005 for assessment of disease dynamics
2. To determine whether growth anomalies affect the growth of table corals
3. To determine whether removal of growth anomalies off of table corals will enhance the growth or reproduction of affected colonies
4. To determine distribution and prevalence of fish disease
5. To determine the affect of skin cancer on the body condition of fish
6. To determine distribution and prevalence of parasitic diseases in zooplankton

**7. Answer the Findings below by providing information that you believe will assist the Co-Trustees in determining how your proposed activities are compatible with the conservation and management of the natural, historic, and cultural resources of the Monument:**

The Findings are as follows:

- a. How can the activity be conducted with adequate safeguards for the cultural, natural and historic resources and ecological integrity of the Monument?

Activities will be conducted in a manner to minimally impact coral reef resources and standard protocols for disease studies developed for the Monument will be used. All gear will be sterilized each day and any collected organisms will be placed in plastic bags at depth before transfer to the small boat. All laboratory work will be conducted using established biosecure protocols including sterilizing all tools and work surfaces. All biological samples will be fixed in solution for transport to our laboratories in Honolulu.

- b. How will the activity be conducted in a manner compatible with the management direction of this proclamation, considering the extent to which the conduct of the activity may diminish or enhance Monument cultural, natural and historic resources, qualities, and ecological integrity, any indirect, secondary, or cumulative effects of the activity, and the duration of such effects?

The collapse of coral reefs from disease in other regions points to the critical need to understand disease processes. Our research program is dedicated to studying disease in the Monument so that managers have the information they need to protect these vulnerable resources. All research proposed in this permit application is directly

applicable to the management of diseases of coral and fish within the region. All surveys are conducted in a manner causing little to no impact on the environment as they use visual and photographic techniques. We will be collecting the minimal number of fish or coral samples required to complete our laboratory analyses.

c. Is there a practicable alternative to conducting the activity within the Monument? If not, explain why your activities must be conducted in the Monument.

There is no alternative to conducting the activity in the Monument. Although, comparative studies of disease in other regions are useful, they cannot replace understanding damage from the specific diseases affecting fish and coral populations in the Monument.

d. How does the end value of the activity outweigh its adverse impacts on Monument cultural, natural and historic resources, qualities, and ecological integrity?

If diseases are not managed in the Monument, the coral reefs will suffer the same fate as coral reefs in the Florida Keys and other regions of the Caribbean. In the Keys, their acroporoids, which used to be their numerically dominant coral, have been reduced by 90% and are now on the endangered species list (Patterson et al. 2002). Acroporoids in the Monument are already in decline due to two different diseases, Acropora white syndrome and Acropora growth anomalies. Current models of global climate change predict a significant increase in sea surface temperature (Kleypas et al. 1999). Elevated temperatures have been shown to accelerate the growth rate and pathogenicity of pathogens and so it is predicted coral disease will become more common and widespread (Porter et al. 2001). On the GBR, increases in White Syndrome are associated with temperature anomalies. Acropora white syndrome is also currently killing corals in the Monument so information of the epizootiology of this disease is critically important for the development of both immediate and long-term management strategies. Reductions in fish populations from overfishing has contributed to the decline of reefs as algae are allowed to outcompete corals. The Monument is closed to fishing but its fish populations are known to suffer from diseases. Kole (*Acanthurus strigosus*), an important herbivore, was found to have skin cancer. However, nothing is known about the distribution or prevalence of this disease in fish populations. Our studies on

coral and fish diseases provide critical information necessary for management to address the already established disease outbreaks degrading the coral reefs of the Monument.

e. Explain how the duration of the activity is no longer than necessary to achieve its stated purpose.

We are requesting the absolute minimum amount of time require to condut our studies.

We anticipate staying a maximun of 5 days at any one island within the Monument.

f. Provide information demonstrating that you are qualified to conduct and complete the activity and mitigate any potential impacts resulting from its conduct.

I have been conducting coral disease surveys and studies in the Monument since 2002.

I am familiar with the reefs and methodology required to safely conduct all proposed studies. I was involved in the development of protocols for investigations of coral disease developed for the Monument. I am also a co-author on the book "A coral disease handbook: guidelines for assessment, monitoring and management." and helped develop Hawaii Division of Aquatic Resources "Rapid Response Contingency Plan for unusual events of coral bleaching, disease and COTS outbreaks". Both of these publications make recommendations for proper procedures involving investigating marine diseases including field techniques, the need for follow-up laboratory investigations and safe handling of samples.

g. Provide information demonstrating that you have adequate financial resources available to conduct and complete the activity and mitigate any potential impacts resulting from its conduct.

I am employed by the University of Hawaii and thus would be covered under University policies.

h. Explain how your methods and procedures are appropriate to achieve the proposed activity's goals in relation to their impacts to Monument cultural, natural and historic resources, qualities, and ecological integrity.

The Monuments goal is to preserve the integrity of the resources. Disease is already established in the Monument and is starting to degrade the acroporid populations. Fish disease has been documented in herbivorous fish in the Monument but there is no information on the extent of the fish populations affected. If the Momument is to prevent irreparable damage from disease it must first have information on the extent and harm from diseases. Our research addresses these needs for the Monument and does so in the most minimally invasive manner as possible. Our

methods are predominately visual surveys which do no harm. Marking of individual colonies is also non-invasive. Samples will be taken of any new diseases encountered not characterized by histology and surgical removal of growth anomalies will be undertaken to test the efficacy of this method for disease control. Collection of diseased fish are required for follow-up laboratory analyses.

i. Has your vessel has been outfitted with a mobile transceiver unit approved by OLE and complies with the requirements of Presidential Proclamation 8031?

yes

j. Demonstrate that there are no other factors that would make the issuance of a permit for the activity inappropriate.

### **8. Procedures/Methods:**

Disease surveys: As possible, re-survey of established sites at FFS, Pearl and Hermes (PHR), Midway (MID) and Kure (KUR) will be done following established protocol. Two 25 m lines will be laid out along the permanent pins. A diver will then swim over the lines during which all corals within one half meter of either side of the transect lines will be identified to specie, counted, and assigned to a size class (0-5cm; 6-10cm; 11-20cm; 21-40cm; 41-80cm; 81-150cm; >150cm.). In the same manner, a second diver will swim over the lines and examine all corals for signs of bleaching or disease. Bleached colonies will be assigned a bleaching category: 0-no bleaching; 1- 10-30%; 2-30-50%; 3-50-100%; 4- 100%; 5-mortality. For corals exhibiting disease, a general description of the condition will be recorded, the coral will be photographed and a specimen will be collected for histopathological examination. All enumerated bleached and diseased corals will also be assigned a size class consistent with the population counts. Individual colonies tagged in 2005 or 2006 will be relocated, remarked and photographed. Any new infected colonies along the transect will be photographed and tagged. Any lost pins will be replaced and any loose pins re-glued.

Acropora growth studies

Colonies of *A. cytherea* with growth anomalies and a nearest neighbor of similar size will be measured (length and width of each tier), photographed (with a ruler) and tagged. They will be re-examined the following year (2011) to look for differences in growth between affected and control colonies. We will be tagging colonies located at our established permanent sites at FFS.

#### Acropora tumor removal

Ten acropora colonies with growth anomalies and five healthy nearest neighbor colonies will be surveyed as described above. Growth anomalies will be removed from five of the colonies. GAs will be bagged at depth and processed onboard the ship for further study. This removal treatment has already been used, to a limited extent, by scientists (G. Williams) at Palmyra Atoll. Although only a few colonies were treated, it was found that growth anomaly removal was effective, there was no evidence of disease spread or that treatment adversely affect the treated colony and was effective in stopping the disease. Although, we do not know whether this disease is infective or not, if it is, removal of the GAs would also remove a potential source of infection from the reefs. In addition, to determine whether growth anomaly removal might also enhance reproduction of the colonies a small sample (5-7cm) will be collected off of all colonies (5 healthy, 5 control GA, 5 treated GA). All colonies will be re-surveyed and sampled the following year in May to determine effect of the treatment on the growth and reproduction of colonies. Surveys will also reveal whether the disease re-occurs after treatment and if so, to what extent. On the ship, GA samples will be fixed in zinc formalin for histology, gluteraldehyde for electron microscopy or bleached for skeletal analysis. Healthy coral samples will be fixed in zinc formalin for histology and polyp dissections.

#### Fish disease studies

##### Surveys

Visual surveys of populations of surgeonfish and butterflyfish will be conducted at all islands visited. Census techniques will consist of linear timed swims (using SCUBA) of 45-60 min where all acanthurids and chaetodontids with and without skin lesions or tumors will be

enumerated. Census tracks will be recorded using a float with GPS carried by one of the divers. Area surveyed will be estimated from the GPS track, and the fish census data will be used to calculate fish density (fish/unit area) and prevalence of fish with skin lesions. These same methods have been used to document prevalence of fish disease within the MHI.

### Histology

Fish with noticeable lesions will be collected by spear, placed on ice and transported to the ship for examination. Fish will be weighed and measured (standard and fork length), examined systematically externally and internally, and gross lesions documented. For histopathology, sections of skeletal muscle, skin, spleen, liver, cranial and caudal kidneys, swim bladder, brain, heart, gill, and gonad, small intestines, and stomach will be excised and fixed in 10% neutral buffered formalin. Tissues will be sectioned, dehydrated in alcohol series, embedded in paraffin, sectioned at 5 µm, placed on microscope slides, stained with hematoxylin and eosin, and examined using a light microscope. Special stains will be used as appropriate to identify fungi, bacteria, or protozoa. Histopathology will allow us to characterize microscopic morphology of disease, will provide systematic evaluation of cellular changes that occur in disease, and will afford the opportunity to detect microorganisms and the host response to these organisms.

### Zooplankton surveys

Zooplankton will be collected, opportunistically, at the end of each workday by doing plankton tows as we return to the ship. We will also, as possible, collect samples using light traps off of the ship in the evenings. All samples will be preserved in 10% formalin and transported back to our lab in Honolulu. Samples will be sorted and screened for parasitic infections.

**NOTE: If land or marine archeological activities are involved, contact the Monument Permit Coordinator at the address on the general application form before proceeding, as a customized application will be needed. For more information, contact the Monument office on the first page of this application.**

### **9a. Collection of specimens - collecting activities (would apply to any activity): organisms or objects (List of species, if applicable, attach additional sheets if necessary):**

Common name:

table coral

Scientific name:

Acropora cytherea

# & size of specimens:

15 samples (5 from healthy, 5 from treated GA tables, 5 from control GA tables) size of sample: 5-7cm

Collection location:

FFS

Whole Organism  Partial Organism

**9b. What will be done with the specimens after the project has ended?**

Samples will be used up in the analyses (polyp dissections)

**9c. Will the organisms be kept alive after collection?**  Yes  No

Samples will be kept alive until they can be fixed in zinc formalin on the ship

• General site/location for collections:

FFS

• Is it an open or closed system?  Open  Closed

• Is there an outfall?  Yes  No

• Will these organisms be housed with other organisms? If so, what are the other organisms?

no

• Will organisms be released?

no

**10. If applicable, how will the collected samples or specimens be transported out of the Monument?**

Transport of preserved samples out of the Monument would occur during transit between islands and back to the MHI

**11. Describe collaborative activities to share samples, reduce duplicative sampling, or duplicative research:**

No other acroporid disease studies are currently underway in the Monument.

**12a. List all specialized gear and materials to be used in this activity:**

dive gear  
coral collection gear (bone cutters, hammer, chisel, ziplock and whirlpak bags, bag to carry gear)  
coral processing gear (plastic jars, z-fix, gluteraldehyde, clorox)  
stereo microscope  
fish dissecting gear (scissors, scalpels, forceps, scale, rulers, plastic jars, formalin)  
cameras and underwater housing  
sludge hammer, steel pins and underwater glue  
field equipment (tape measures, floats, clipboards, underwater paper, cow ear tags, cable ties)  
hand held GPS  
computer  
5 gal buckets with lids  
plankton net  
zooplankton light trap  
Miscellaneous office supplies (books, tablets, pencils, pens, markers, scissors, stapler, 3-hole punch, etc.)  
Personal gear (clothing, personal hygiene items, diet coke, snacks, sunglasses, etc)

**12b. List all Hazardous Materials you propose to take to and use within the Monument:**

Clorox- Sodium Hypochlorite- irritant-5 gallons-used for sterilization of equipment and growth anomaly processing for skeletal analyses.  
Z-fix-zinc formaldehyde-irritant-1gallon-used for preserving coral samples for histology  
Ethanol-ethyl alcohol-flammable-1 gallon-used for preserving samples for molecular analyses  
Gluteraldehyde-corrosive-1 gallon-used for preserving coral samples for electron microscopy  
Formaldehyde-irritant-1 gallon- used for preserving fish samples for histology  
All chemicals will be contained in bottles within secondary containment and will be transported out of the Monument and sent back to our lab in Honolulu

**13. Describe any fixed installations and instrumentation proposed to be set in the Monument:**

: Repair or replacement of steel pins at permanent monitoring sites.

**14. Provide a time line for sample analysis, data analysis, write-up and publication of information:**

Fall 2010: histology, polyp dissections and parasitology processing. Spring -Fall 2011: data analysis and report writing

**15. List all Applicants' publications directly related to the proposed project:**

Aeby, G, Work, T, Fenner, D. and E. DiDonato. 2009. Coral and crustose coralline algae disease on the reefs of American Samoa. Proc Int. 11th Int. Coral Reef 197-201

Work, T., Aeby, G., Stanton, F., and D. Fenner. 2008. Overgrowth of fungi (endolithic hypermycosis) associated with multifocal to diffuse distinct amorphous dark discoloration of corals in the Indo-Pacific. Coral Reefs 27:663.

Work, T., Aeby, G. and S. Coles. 2008. Distribution and morphology of growth anomalies in Acropora from across the Indo-Pacific. Dis. Aquat. Org. 78(3):255-264.

Williams, G., Davy, S. and G. Aeby. 2008. Coral disease at Palmyra Atoll, a remote reef system in the Central Pacific. Coral Reefs 27:207.

Kenyon, J., Dunlap, M., Wilkinson, C. Page, K., Vroom, P and G. Aeby. 2007. Community structure of hermatypic corals at Pearl and Hermes Atoll, Northwestern Hawaiian Islands: Unique conservation challenges within the Hawaiian archipelago. Atoll Research Bulletin 549.

Kenyon, J., Wilkinson, C., Dunlap, M., Aeby, G. and C. Kryss. 2007. Community structure of hermatypic corals at Laysan and Lisianski/Neva Shoal, Northwestern Hawaiian Islands: A new layer of scientific exploration. Atoll Research Bulletin 550:

Aeby, G.S. 2007. First record of coralline lethal orange disease (CLOD) in the Northwestern Hawaiian Islands. *Coral Reefs* 26(2):385.

Aeby, G.S. 2007. Spatial and temporal patterns of infection of *Porites* trematodiasis on the reefs of Kaneohe Bay, Oahu, Hawaii. *Bull. Mar. Sci.* 80(1):209-218.

Aeby, G.S. 2006. Baseline levels of coral disease in the Northwestern Hawaiian Islands. *Atoll Research Bulletin* 543:471-488.

Domart-Coulon, J., N.Traylor-Knowles, E. Peters, D. Elbert, C. Downs, K. Price, J. Stubbs, S. McLaughlin, E. Cox, G. Aeby, P. Brown and G. Ostrander. 2006. Comprehensive characterization of skeletal tissue growth anomalies of the finger coral *Porites compressa*. *Coral Reefs* 25:531-543. Symp. 197-201.

Kenyon, J., Wilkinson, C. and G. Aeby. 2008. Community structure of hermatypic corals at Maro reef in the Northwestern Hawaiian Islands: A unique open atoll. *Atoll Research Bulletin* 558.

Work, T. and G. Aeby. 2006. Systematically describing gross lesions in corals. *Dis Aquatic Org* 70:155-160.

Kenyon, J. G. Aeby, R. Brainard, J. Chojnacki, M. Dunlap, C. Wilkinson. 2006. Mass coral bleaching on high-latitude reefs in the Hawaiian Archipelago. *Proc. 10th Int. Coral Reef Symp.* 631-643.

Kenyon, J., Vroom, P., Page, K., Dunlap, M., Wilkinson, C. and G. Aeby. 2006.

Community Structure of Hermatypic Corals at French Frigate Shoals, Northwestern Hawaiian Islands: Capacity for Resistance and Resilience to Selective Stressors. *Pac Sci* 60(2):153-175.

Maragos, J., D. Potts, G. Aeby, D. Gulko, J. Kenyon, D. Siciliano, and D. VanRavensway. 2004. The 2000-2002 Rapid Ecological Assessment of Corals in the Northwestern Hawaiian Islands, Part I: Species and Distribution. *Pacific Science* 58(2):211-230 .

Aeby, G. 2003. Corals in the genus *Porites* are susceptible to infection by a larval trematode. *Coral Reefs* 22:216.

Aeby, G.S., Kenyon, J., Maragos, J. and Potts, D. 2003. First record of mass coral bleaching in the Northwestern Hawaiian Islands. *Coral Reefs* 22:256.

#### Literature cited

Aeby, G.S. 2006a. Baseline levels of coral disease in the Northwestern Hawaiian Islands. *Atoll Research Bulletin* 543:471-488.

Aeby, G.S. 2006b. Outbreak of coral disease in the Northwestern Hawaiian Islands. *Coral Reefs* 24(3):481.

Aeby, G. S., Kenyon, J., Maragos, J. and Potts, D. 2003. First record of mass coral bleaching in the Northwestern Hawaiian Islands. *Coral Reefs* 22:256.

Aronson, R. B. and W. F. Precht. 2001. White-band disease and the changing face of Caribbean coral reefs. *Hydrobiologia*. 460: 25-38.

Barber, R., A. Hilting, and M. Hayes. 2001. The changing health of coral reefs. *Human and Ecological Risk Assessment*: 7(5):1255-1270.

Brown, B. 1997. Coral bleaching: causes and consequences. *Coral Reefs* 16:S129-S138.

Friedlander AM, Parrish JD, DeFelice RC (2002) Ecology of the introduced snapper *Lutjanus kasmira* (Forsskal) in the reef fish assemblage of a Hawaiian Bay. *J Fish Biol* 60:28-48

Green, E. and Bruckner, A. 2000. The significance of coral disease epizootiology for coral reef conservation. *Biological Conservation*. 96: 347-361.

Hoegh-Guldberg, O. 1999. Climate change, coral bleaching and the future of the world's coral reefs. *Marine Freshwater Research* 50:839-866.

Kenyon, J.C., Aeby, G., Brainard, R., Chojnacki, J., Dunlap, M. and C. Wilkinson 2006. Mass coral bleaching on high-latitude reefs in the Hawaiian Archipelago. *Proceedings of the 10th Int. Coral Reef Symposium, Okinawa*. pp. 631-643.

Peters, E. 1997. Diseases of coral reef organisms. In: Birkeland, C. (Ed.). *Life and Death of Coral Reefs*. Chapman & Hall, London, pp.114-136.

Porter, J., P. Dustan, W. Jaap, K. Patterson, V. Kosmynin, O. Meier, M. Patterson, and M. Parsons. 2001. Patterns of spread of coral disease in the Florida Keys. *Hydrobiology* 159: 1-24.

Randall JE (1987) Introduction of marine fishes to the Hawaiian Islands. *Bull Mar Sci* 41:490- 502

Richardson, L. 1998. Coral diseases: what is really known? *Trends in Ecol. Evol.* 13 (11):438-443.

Santavy, D., Peters, E. 1997. Microbial pests: Coral disease in the Western Atlantic. *Proc 8th Int Coral Reef Sym* 1:607-612.

Santavy, D., Mueller, E., Peters, E., MacLaughlin, L., Porter, J., Patterson, K. & Campbell, J. 2001. Quantitative assessment of coral diseases in the Florida Keys: strategy and methodology. *Hydrobiologia.* 460: 39-52.

Walker, H. 2001. Understanding and managing the risks to health and environment from global atmospheric change: A synthesis. *Human and Ecol Risk Assessment* 7(5):1195-1209.

Work T, Rameyer RA, Takata G, Kent M. 2003. Protozoal and epitheliocystis-like infections in the introduced blueline snapper *Lutjanus kasmira* in Hawaii. *Diseases of Aquatic Organisms* 37:59-66.

With knowledge of the penalties for false or incomplete statements, as provided by 18 U.S.C. 1001, and for perjury, as provided by 18 U.S.C. 1621, I hereby certify to the best of my abilities under penalty of perjury of that the information I have provided on this application form is true and correct. I agree that the Co-Trustees may post this application in its entirety on the Internet. I understand that the Co-Trustees will consider deleting all information that I have identified as “confidential” prior to posting the application.

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Signature

Date

**SEND ONE SIGNED APPLICATION VIA MAIL TO THE MONUMENT OFFICE BELOW:**

Papahānaumokuākea Marine National Monument Permit Coordinator  
6600 Kalaniana'ole Hwy. # 300  
Honolulu, HI 96825  
FAX: (808) 397-2662

**DID YOU INCLUDE THESE?**

- Applicant CV/Resume/Biography
- Intended field Principal Investigator CV/Resume/Biography
- Electronic and Hard Copy of Application with Signature
- Statement of information you wish to be kept confidential
- Material Safety Data Sheets for Hazardous Materials

**Supplement for collection**  
**Aeby**

Common name: reef fish

Scientific name: Targeted fish species include *Lutjanus kasmira*, *Ctenochaetus strigosus*, *Acanthurus nigrofuscus*, *Acanthurus sp.*, *Mulloidichthys vanicolensis*, *M. flavolineatus*, *Parupeneus multifasciatus*, *P. pleurostigma*, and *M. pflugeri*

# & size of specimens: We will collect a maximum of 20 fish per specie per island for *Lutjanus kasmira* and the five goatfish species. For affected surgeonfish, *Ctenochaetus strigosus*, *A. nigrofuscus* or other species with lesions, we are requesting a maximum of 10 affected and 10 healthy fish per specie per island.

Collection location: shallow water reefs throughout Monument

x Whole Organism Partial Organism

**1b. What will be done with the specimens after the project has ended?** The fixed samples will be transported to Oahu for parasitological and histopathological analysis.

**1c. Will the organisms be kept alive after collection?** Yes x No

• Specific site/location:

• Is it an open or closed system?  Open X Closed

• Is there an outfall?  Yes XNo

• Will these organisms be housed with other organisms? If so, what are the other organisms? no

• Will organisms be released? no

**2. If applicable, how will the collected samples or specimens be transported out of the Monument?** Samples will be transported on ice on small boats to the Hi'ialakai which may or may not be within Monument waters. Fixed samples will be transported back to Honolulu via the Hi'ialakai.

**3. Describe collaborative activities to share samples, reduce duplicative sampling, or duplicative research:** Fish samples will be collected by and shared with Brian Bowen's group and so they will also be used for both molecular and life history studies.

Common name: reef coral

Scientific name: *Porites sp.*, *Montipora sp.*, *Pocillopora sp.*, *Pavona sp.* Species will vary depending upon disease occurrence.

# & size of specimens: We will only collect samples of diseased corals that have not already been characterized by histology. We anticipate a maximum of 30 samples (all islands combined) to be collected if new diseases are encountered. Sample size would be 2-5 cm each. 2 samples would be taken per colony (one from the diseased region and one from the healthy region).

Collection location: shallow water reefs within the Monument  
Whole Organism  Partial Organism

**1b. What will be done with the specimens after the project has ended?** Fixed samples will be transported to Oahu for histopathological analyses.

**1c. Will the organisms be kept alive after collection?** Yes  No

However, please note that corals will be transported live in buckets of sea water to the Hi'ialakai where they will be placed in Z-fix.

• Specific site/location:  
• Is it an open or closed system?  Open  Closed

• Is there an outfall?  Yes  No

• Will these organisms be housed with other organisms? If so, what are the other organisms? No

• Will organisms be released? No

**2. If applicable, how will the collected samples or specimens be transported out of the Monument?** Samples will be transported in buckets on small boats to the Hi'ialakai which may or may not be within Monument waters. Fixed samples will be transported back to Honolulu via the Hi'ialakai.

**3. Describe collaborative activities to share samples, reduce duplicative sampling, or duplicative research:** none

Common name: Zooplankton

Scientific name: Will vary by sample

# & size of specimens: 100s-1000s of zooplankton

Collection location: shallow water reefs throughout Monument  
 Whole Organism  Partial Organism

**1b. What will be done with the specimens after the project has ended?** The fixed samples will be transported to Oahu for parasitological analysis.

**1c. Will the organisms be kept alive after collection? Yes x No**

- Specific site/location:
- Is it an open or closed system?  Open X Closed
- Is there an outfall?  Yes XNo
- Will these organisms be housed with other organisms? If so, what are the other organisms? no
- Will organisms be released? no

**2. If applicable, how will the collected samples or specimens be transported out of the Monument?** Samples will be transported in buckets on small boats to the Hi'ialakai which may or may not be within Monument waters. Fixed samples will be transported back to Honolulu via the Hi'ialakai.

**3. Describe collaborative activities to share samples, reduce duplicative sampling, or duplicative research:** no other zooplankton studies are currently underway in the Monument