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
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: Charles Littnan
Affiliation: NOAA Fisheries

Permit Category: Research
Proposed Activity Dates: June 25, 2011- July 31, 2012
Proposed Method of Entry (Vessel/Plane): NOAA RV O.E. Sette, possibly a chartered vessel (e.g. the Searcher) or chartered airplane as necessary.
Proposed Locations: Kure Atoll, Midway Atoll, Pearl and Hermes Reef, Laysan, Lisianski, French Frigate Shoals, Nihoa

Estimated number of individuals (including Applicant) to be covered under this permit: 10
Estimated number of days in the Monument: 15 weeks

Description of proposed activities: (complete these sentences):

a.) The proposed activity would…
consist of efforts to help increase juvenile monk seal survival in the Northwestern Hawaiian Islands.

b.) To accomplish this activity we would ….
undertake enhancement actions including: 1) feeding and treating prematurely weaned and other undernourished seals in a captive facility, 2) treating weaned/juveniles to decrease parasite loads.

c.) This activity would help the Monument by …
determining effective methods to aid in the recovery of endangered Hawaiian monk seal and thus maintain biodiversity within the Monument.

Other information or background:

Section A - Applicant Information

1. Applicant

Name (last, first, middle initial): Littnan, Charles L.

Title: Leader, Hawaiian Monk Seal Research Program
Pacific Islands Fisheries Science Center, NOAA Fisheries

1a. Intended field Principal Investigator (See instructions for more information):
Charles L. Littnan
Kathleen Gobush

2. Mailing address (street/P.O. box, city, state, country, zip): [redacted]

Phone: [redacted]

Fax:

Email: [redacted]

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

3. Affiliation (institution/agency/organization directly related to the proposed project):
Pacific Islands Fisheries Science Center/NOAA Fisheries/Department of Commerce

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):
Frances Gulland- Veterinarian
Chad Yoshinaga- field researcher
Jessie Lopez- field researcher
Angie Kaufman- field researcher
TBD veterinarian
TBD field research staff

Section B: Project Information

5a. Project location(s):

		Ocean Based	
☒ Nihoa Island	☒ Land-based	☒ Shallow water	☐ Deep water
☐ Necker Island (Mokumanamana)	☐ Land-based	☐ Shallow water	☐ Deep water
☒ French Frigate Shoals	☒ Land-based	☒ Shallow water	☐ Deep water
☐ Gardner Pinnacles	☐ Land-based	☐ Shallow water	☐ Deep water
☐ Maro Reef			
☒ Laysan Island	☒ Land-based	☒ Shallow water	☐ Deep water
☒ Lisianski Island, Neva Shoal	☒ Land-based	☒ Shallow water	☐ Deep water
☒ Pearl and Hermes Atoll	☒ Land-based	☒ Shallow water	☐ Deep water
☒ Midway Atoll	☒ Land-based	☒ Shallow water	☐ Deep water
☒ Kure Atoll	☒ Land-based	☒ Shallow water	☐ Deep water
☐ Other			

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

Location Description:

Location will be the sand beaches and near shore habits of all islands specified, including sand beaches of all islets within the atoll complexes of Kure Atoll, Midway Atoll, Pearl & Hermes Atoll, and French Frigate Shoals. Most of the effort, however, will take place in these habitats at French Frigate Shoals, Laysan Island, Lisianski Island, and Nihoa Island.

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:***
The Hawaiian monk seal is on a path to extinction that is unlikely to be altered without human intervention. This application presents tools for improving the survival of young seals, an essential requirement for averting extinction.

The total abundance of Hawaiian monk seals in the Northwestern Hawaiian Islands (NWHI), has declined by 70 % since the late 1950s. Since then, the six main sub-populations have experienced everything from periods of promising growth to catastrophic setbacks. The causes of decline have varied over time and from place to place, but since the early 1990s the decline has been driven, in large part, by poor juvenile survival. Many of these young animals have failed to thrive, and only about 1 of every 5 live to reach maturity, a situation largely due to insufficient food availability. The age structure of the population is therefore now unfavorable for future growth and the total population will inevitably fall below 1,000 individuals in just a few years.

The decline will continue and the conservation challenge will intensify unless scientists and managers, working together, develop the means to improve juvenile survival. History teaches us that the monk seal will continue to face new and unforeseen challenges in the future, but after two decades of poor juvenile survival, it is clear that this problem must be addressed. Improving juvenile survival is one of four key activities highlighted in the new Recovery Plan for the Hawaiian monk seal, published by NOAA in the summer of 2007:

- Improving juvenile survival through direct intervention such as providing captive care and feeding;
- Mitigating mortality due to entanglement in marine debris;
- Reducing shark predation on seal pups; and
- Ensuring growth of the small Main Hawaiian Islands seal population.

All of these critical recovery activities are being pursued by NOAA and its partners. The work proposed here focuses on direct interventions with juvenile seals to improve their survival and thus better position the monk seal population for recovery in the future.

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?

The ultimate goal of the work described here is to assist in the recovery of the Hawaiian monk seal, a goal that is consistent with Monument mandates. The research proposed herein is

compatible with the conservation and management goals of the Monument and minimizes disturbance to the NWHI ecosystem.

Our studies will be designed and executed so as to minimize impacts to the terrestrial and marine environment. For instance, on-island time will be limited to that required for animal capture, transport, and instrument deployment, during which all personnel will adhere to strict quarantine protocols as defined by USFWS. Movements will be confined to the immediate beach area to avoid potential disturbance to bird and plant life on the island interiors. After the final tag deployment or adequate monitoring period, NMFS monk seal researchers will arrange to return to the NOAA R/V Oscar Elton Sette or other vessel, thereby reducing any human disturbance to terrestrial habitats and species by returning early.

Native Hawaiians share a close link to the ocean, marine life, and islands within the monument and seek to maintain the living cultural resources found there. Hawaiian monk seals are one of the most threatened of these cultural and natural legacies. The work presented here is critical for the survival of this species into the future, and it is our intent to continue this work with respect and in partnership with the Native Hawaiian community, Accordingly, all scientists participating on these cruises will receive a Native Hawaiian cultural briefing before departure to the NWHI. In addition, the primary permittee, chief scientist, and other appropriate personnel look forward to consulting with the Office of Hawaiian Affairs (OHA) and the Monument's Native Hawaiian program coordinator on proper conduct while in the NWHI, on cultural sensitivities associated with the proposed activities and locations, and on the applicability of the results of this research to the role of OHA as one of the NWHI stakeholder agencies.

Instruments that may be deployed on seals include: wildlife computers spot 5 location only satellite tags and/or mark10 GPS satellite tags.

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?
Please see 7a.

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.
The techniques proposed here to improve juvenile survival can only be applied to seals in the NWHI. This population, unlike seals in the MHI, is demonstrating a population decline and nutrionally stressed seals.

d. How does the end value of the activity outweigh its adverse impacts on Monument cultural, natural and historic resources, qualities, and ecological integrity?
The potential gain from this project is the increased survival of juvenile monk seals in the rapidly dwindling NWHI population. This work if successful and applied on a broader scale in the future could slow or stop the population decline.

e. Explain how the duration of the activity is no longer than necessary to achieve its stated purpose.
All activities here are devised in a manner to minimize time in the field. Researchers will remain in the field for only the time necessary to handle, treat and monitor seals to a degree that ensures the success of the studies and actions proposed here. Most work proposed here is also intended to occur in conjunction with population assessment camps already in place.

f. Provide information demonstrating that you are qualified to conduct and complete the activity and mitigate any potential impacts resulting from its conduct.
The Hawaiian Monk Seal Research Program has been conducting research on this species for over two decades. All members participating on these studies have previous monk seal handling experience. The protocols and research plans presented for these studies have been reviewed and approved by a variety of experts including the Marine Mammal Commission, Hawaiian Monk Seal Recovery Team, as well as other external specialists.

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.
All research/enhancement activities are supported by NOAA Fisheries funding and primarily with the use of NOAA research vessels.

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.
All participating staff are educated and trained to respect all cultural, natural and historic resources in the Monument. Our first and primary objective is "Do no harm". See section 7a above for details.

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.

There are no factors, such as other permit violations, that should prevent the issuance of this permit. All activities are inline with Hawaiian Monk Seal Recovery Plan and relevant sections of the Monument Management Plan.

8. Procedures/Methods:
A range of prospective approaches for increasing juvenile survival have been identified, including:
• Bringing young animals into captivity for feeding and veterinary care, followed by release back into the wild
• Treatment of free-ranging young animals to reduce parasite loads

Based on past experience, scientific review, and detailed consultations with external specialists, these two general approaches have been identified as the interventions most likely to be successful. Some latitude is required in the application of these interventions because at any given time, the optimal approach will depend on a number of factors such as the relative survival among the different sites, the logistics of moving animals, the availability of favorable release sites and so on.

Captive Feeding Program

The Hawaiian monk seal program will collect, as appropriate, juvenile seals (0-3 years old) to feed, treat, and protect in captivity. Seals selected for this work will be those that are prematurely weaned, undernourished twins, or otherwise in a condition that without captive care and supplemental feeding will perish. Seals will be cared for in shore pens (see shore pen description below) or transported to and cared in the main Hawaiian Islands with the intent to release them back at their natal site in the NWHI. Captive care operations will be dependent on the Ford Island Facility and/or the NELHA monk seal rehabilitation center coming on-line in 2011.

On-site operations.
Field operations will be needed to assess, capture, and hold animals that would benefit from interventions to improve their survival. Assessment of individual seals is a routine element of ongoing annual studies. Capturing seals is more complicated because juvenile animals, in particular, may be absent from the islands for weeks at a time. Therefore, on-site holding is almost always required because capture cannot be reliably timed to coincide with the arrival and departure of a transport vessel or aircraft (see description of shore pen below).

Transport of animals from NWHI to MHI and return.
As much as possible, captive care operations will be supported by existing vessel and aircraft activity associated with establishing and retrieving annual field camps. In the past, the U.S. Coast Guard, U.S. Navy, and U.S. Air Force have provided additional assistance opportunistically, and similar arrangements will be sought to minimize transportation costs. In spite of such welcome help, additional chartering of both vessels and aircraft may be necessary.

Release and post-release monitoring.
Releasing animals may require temporary holding facilities if a “soft release” method (i.e, gradual introduction to the release site) is used. The primary holding pens will measure approximately 30 ft x 80 ft but ultimately the dimensions will be established based on the beach topography and near shore bottom contour. Approximately 30% of the surface area will include water at least 24 inches deep at lowest tide. Approximately 70% of the pen would be dry resting area with sufficient area above the high water surge line. Construction of the pens at the primary site will comprise plastic and/or metal fencing material supported by 10-12 ft tall, 2" diameter steel pipe driven into the sand at approximately 6–8 ft apart in the water and “T posts” at 10-12 ft intervals on shore. Plastic ties will fix the fencing to the upright supports. Windbreaks will be erected along the fence near some dry resting area. Fence perimeters (in and out of water) will be monitored twice daily, and will be repaired or changed as necessary to prevent escape or

injurious entrapment. Location of pens will be dependent on the island, season, local conditions and number of seals to be returned at one time.
Staff will be needed to provide care at the release site, as well as to release and monitor the animals' acclimation. Monitoring will involve observational assessment to gauge animal condition and health, as well as tracking movement and foraging patterns using well-established tagging technology. In many, if not most, cases, releases will be timed to take advantage of personnel, equipment, and support from concurrent field studies.

As was done with the Midway twin juvenile seals in 2006, any seals under captive care will be returned as soon as it is advised by the attending veterinarian, the individual has passed a disease screening and suitable transport back to their natal site is available. We will endeavor to release seals via a soft release program, using shoreline pens, continued monitoring, and supplemental feeding, as well as medical care if necessary. The specifics of this release program will depend on the size, health, stress response of the seal, the number of days it was in captivity and the cause for its being taken into captivity, the season it is returned to its natal site and local conditions, as well as staff availability. Flexibility in the release program is necessary to ensure the best chances of survival upon release. For example, a seal that is released as a younger juvenile may need more supervision upon release than an older, more robust seal. A larger juvenile may need a larger shore pen to mitigate the chance of a stress response developing. A seal that takes a long time to learn to free feed might need a longer period of monitoring and supplemental feeding in a shore pen prior to complete release.

We intended to return seals back to their natal sites, with the following exceptions: 1) extenuating circumstances exist, such as an unusual mortality event is occurring at the natal site at that time, or 2) the natal site is Midway. Juvenile survival has been abysmal at Midway, thus return their would be akin to a death sentence; therefore, a Midway juvenile brought into captive care would be returned to its natal island complex, likely Pearl and Hermes.

For the special case of Nihoa, that has limited beach and beach access, seals would be released from small boat off shore, but as close to the shore break as possible. The seals will be oriented toward the beach (main haul out area) and surrounding ledge. The objective will be to have the seals get on land, rather than swim in the opposite direction toward deeper seas.

Worming
Objective:

Hawaiian monk seal abundance is declining due to low juvenile survival, which appears to be associated with food limitation and poor body condition. Monk seals are known to host a variety of gastrointestinal parasites (Dailey et al. 1988, 2004). Reif et al. (2006) reported that young seals infected with Diphyllobothrium spp. (tape worms) tended to be in poorer body condition than those uninfected, and proposed that "intervention strategies to reduce the gastrointestinal helminth burdens in immature animals should be considered as a conservation measure." Our study is designed to test the hypothesis that temporarily relieving compromised young monk seals of their parasite burden will improve their chances of survival in a food limited environment. Specifically we will test the potential for enhanced survival of seals, aged young-

of-the-year to 2-years old, following treatment to reduce gastro-intestinal parasite load.

Experimental design and protocols:

Selection of subject animals (pre-treatment assessment)
The study will focus on NWHI juvenile seals up to two years of age which corresponds to the age range exhibiting the lowest survival and which is primarily responsible for constraining population growth and recovery. Sample sizes will be limited by the number of juveniles available at each study site which match the specified selection criteria (see below), or by the capability of the field teams to identify, capture and treat the seals safely in the allotted time. The objective will be to include all available candidate seals in the study. Based on cohort numbers over the last 5 years, the maximum number of seals in each age class (0-2) that may be included in the study each year are: French Frigate Shoals: 47 seals; Laysan Island: 41 seals; and Lisianski Island: 29 seals. Standard population surveys will be conducted to identify potential study subjects. We will attempt to treat all animals that fall into the target age class. If the opportunity to treat seals is limited in certain cases and in order to test the hypothesis above, we will prioritize sampling for animals that are most likely to be compromised by nutritional stress and parasites, but which are not moribund and unlikely to survive under any circumstances.

Pre-treatment assessments of health status and body condition will be based on visual inspection, supplemented by examination of digital photos. The monk seal program employs a suite of body condition indices to score seals as good/medium/thin/emaciated. These indices rely primarily on the relative visibility of: “pelvic girdle” (ischium, greater trochanter of the femur, ilium), ribs, point of the shoulder (scapulohumoral joint), peanut head (circumferential loss of mass around the neck and cranial shoulders), and vertebrae (dorsal spinous processes and transverse processes). Seals scored as “emaciated” often disappear from the population soon thereafter or before the start of the subsequent field season, and if it is felt that they are too compromised to treat without severe risk of mortality they will be excluded from this study.

Controls
Seals will be assigned to either a treatment or control group using either random assignment. The objective will be to obtain an equivalent number of seals in both treatment and control groups, matched as closely as possible in age, sex, body condition, and location. Sex matching is important because sex has been recognized to influence worm burden and its effects on the host in other mammals (Wilson and Moore 2002).

Location and Timing
Historically, juvenile survival has varied markedly both temporally and spatially (Baker and Thompson 2006). Further, the pattern of the relationship between pup condition (weaning girth) and survival also varies annually, apparently due to environmental stochasticity (Baker et al. 2007), predation intensity and other factors (Baker 2008).

This study has been conducted at Laysan Island since August 2009. To date, 43 juveniles seals have been apart of the study. Results analyzed thus far have been equivocal but show promise. For example, seals treated with injectable Praziquantel gained more mass on a daily basis in the

period of March to May (2010) as compared to control seals; however at other time periods throughout the year, the two groups of seals were indistinguishable. In order to conclusively test our hypotheses, we aim to conduct additional trials on additional juveniles seals, possibly using different routes (i.e. topical versus oral and injectable), dosages (because drug dose is often route-dependent) and possibly additional drugs (i.e. nematodicides).

Drugs and routes tried have included oral Fenbendazole, a nematodicide (10-50 mg/kg), and oral and injectable Praziquantel, a cestodicide (5 mg/kg). Approval for use of injectable ivermectin, a nematodicide (0.02 mg/kg) has been granted but we have yet to use it. We may try it in 2011 as well as a new anti-helminthic drug, just now commercially available called Profender ® Topical Solution (Bayer, Inc.). Profender® combines emodepside, a nematodicide, (suggested dose 3 mg/kg but may be used at a dosage approximately 3 times as high to be effective in pinnipeds) and praziquantel, a cestodicide (suggested dose 12 mg/kg but may be used at a dosage approximately 3 times as high to be effective in pinnipeds) a highly effective treatment for roundworms, tapeworms and hookworms in cats.

For 2011, in order to detect and describe the effects of the treatment, the study may be conducted at multiple sites and with multiple anti-helminth medications and routes of administration (a s listed above). Laysan Island will serve as one of the study sites because there is a relatively large number of pups born annually and apparently a minimum of mortality factors other than food limitation (e.g., predation) to confound the results. Also, because it is a single island, all animals on the beach on any given day are available for treatment and observation (as opposed to the greater logistic difficulties at multi-islet atolls). Additional study sites may include Lisianski Island and French Frigate Shoals. Lisianski is similar to Laysan in terms of known mortality causes and physiography. FFS may serve as an additional site if funding and logistics prove favorable to extending the study.

Seals will be handled for treatment or assessment (sampling or weighing) up to eight times each year. Seals age 1-2 will be treated during any of these times. Pups of the year will be treated once they have been weaned for at least 120 days. The constraint is based on results from epidemiological sampling conducted 1998-2000 (n=54 for weaned pups), which indicated that all pups sampled more than 120 days post-weaning (n=15) tested positive for either cestodes or nematodes (Appendix A: Figure 1). Conversely, only one pup sampled at less than 75 days post-weaning (n=39) tested positive.

Treatment and Assessment Protocols
All study subjects will be captured by hand and net, feces collected for subsequent determination of parasite burden/presence (voided feces or fecal sample collected via fecal loop or digital extraction; stored in 10 % formalin), measured (axillary girth and weight), tagged if necessary, and given a dose of a cestodicide and/or a nematodicide. The cestodicide will be one of the following: Praziquantel oral (Droncit, Bayer 5-10 mg/kg), injectable (Praziquantel injectable, Bayer, 5-10 mg/kg) or topical (Profender, 12-30 mg/kg). The nematocide will be one of the following: oral Fenbendazole (Pancur,10-50mg/kg) or injectable ivermectin (Ivomec 0.2 mg/kg) or topical emodepside (Profender, 3mg-7.5 mg/kg). The drug Profender contains both a cestodicide and a nematocide in one compound.

All anti-helminth medications will be administered in accordance with the protocols developed for elephant seals and California sea lions at The Marine Mammal Center, applied to monk seals during captive rearing studies at Midway Atoll in 2006-7 and captive monk seals at Sea World in San Antonio, Texas and Longs Marine Laboratory, Santa Cruz, California. Control seals will be handled and sampled in the same manner as treatment seals. No sedation will be required for either treatment or control seals. However, the study may be facilitated by conducting it in conjunction with other research involving capture and handling of juvenile monk seals (e.g., foraging and health screen studies), in which case treatments may involve sedation, biomedical sampling, and instrumentation. Post treatment condition and fecal egg pretense and/or counts will be determined by observing the seals, and, when possible, collecting multiple voided fecal samples from known individuals as part of our standard patrols.

We may attempt to re-deworm all treatment seals at approximately 3-16 week intervals or during each subsequent field season. Treatment interval will be dictated by funding, logistics and the medication route tested; for example when testing the topical drug Profender, retreatment may occur on a shorter time interval because handling required is expected to minimal for drug application. The additional treatments serve to clear both the adult worms that survived the previous treatments (or acquired thereafter), as well as migrating larva that matured after the previous treatment. As with the initial treatment, control seals will be handled and measured in the same manner as treatment seals. We will attempt to collect fecal samples from all handled seals. Visual assessment of condition will be recorded on an ongoing basis throughout the study, using standard MMRP subjective body condition scoring (med/thin/emaciated with +/- designations) and scat samples will be collected and preserved for detection of parasites. Subsequent survival will be determined through visual re-identification during regular monk seal population assessment field research, (typically June through August), supplemented by observations made during the additional field sessions for this or other projects. The duration of the survival period will be dependent upon the timing of the initial field phase of the study relative to the assessment field season. That is, at those sites lacking a constant field presence, seals that are not observed in the subsequent field session may have died at anytime during the interim from the previous observation. The resolution of the survival assessment is therefore limited by the frequency of the field presence.

Evaluation:

The evaluation of treatment effects will address two questions:
1. Does treatment of young seals improve their physiological condition (weight, body condition ranking, and parasite load or presence)?
2. Is improved physiological condition (#1) sufficient to increase survival under favorable environmental conditions?

The statistical analysis may consist of modeling survival (either with capture-recapture or logistic regression) of treatment and control animals to determine whether there is evidence that anti-helminth treatment improves survival if there is an apparent difference in survival (as tested initially with a Fisher's exact test). Other factors that influence survival (predation or other) will

be treated as covariates. However, our ability to identify and quantify the underlying environmental factors that drive juvenile survival rates is limited.

Although survival is the ultimate measure, it is a function of a number of factors (physiological condition, environmental conditions, shark predation, entanglement, and other), and worming may not be sufficient to boost survival in all cases. Under severe environmental conditions, animals might show a temporary or seasonal improvement in condition but ultimately not survival. However, that same increase will be made to measure treatment and control seals between each treatment session under more moderate environmental conditions. Consequently, another important analysis will be a comparison of body condition change in treated versus control animals. This assessment may use both quantitative measures (primarily mass) and qualitative measures (MMRP categorical body condition ranking). It should be noted that we might observe different patterns in the effect of worming treatment on seals depending on their life stage. Young of the year are likely to lose weight regardless of treatment as they lose their excess blubber and learn to forage, in this case we would hope that treated seals would show a less precipitous decline in mass and condition than non-treated individuals. Older juvenile seals would be expected to do well to maintain condition and demonstrate some weight gain.

Another key analysis is the comparison of parasite loads in control versus treatment seals. Parasite load will be determined from fecal egg presence or count data, treated as a categorical covariate. Fecal egg counts are not an exact measure of the number of worms present in the stomach and intestine of live animals, as egg production by female worms is influenced by host immunity and worm burden (Gulland and Fox 1992, Zhong and Dobson 1996; Aumont et al. 2003). Furthermore, egg release by tapeworms is intermittent (Reif et al. 2006), whereas nematode eggs are usually randomly scattered in the stools, even if introduced into the fecal stream above the lower colon at irregular intervals as much as several hours apart (Martin 1965; LeJambre et al. 2007). Evaluation of a variety of gastrointestinal nematodes in a number of species has shown that despite variability in fecundity, the fecal egg count is sufficiently repeatable within an individual to allow its use as a quantitative measure of worm burden, and to allow comparisons between individuals (Coltman et al. 1999). Reduction of fecal egg count is the most widely used method to assess the efficacy of anthelminthics against gastrointestinal strongyles (Cabaret and Berrag, 2004). Thus we will use the fecal egg presence and count to evaluate worm burden. Parasite load in any dead animals collected during the study will be determined through an absolute worm count.

Additional descriptions of anchoring:
Anchoring a vessel: small boats may be anchored at any location according to standard practices included in the monk seal field camp permitted activities in order to facilitate transport of seals as necessary. This includes anchoring only in sandy substrate and taking steps to avoid damaging of hard substrates (especially coral) with the anchor or chain.

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:
Hawaiian monk seal

Scientific name:
Monachus schauinslandi

& size of specimens:
Captive Care
An unknown number of seals, but likely less than 10, may be brought to the MHI for captive care. During this time they will be biomedically sampled multiple times to monitor their health and condition.

Worming Trial
Up to 90 juvenile seals (ages 4 months - 2 years) will be captured, sampled and treated for the parasite study. Of these 45 will be controls that will be sampled and 45 treatment seals will be sampled and treated with anti-helminth drugs. Up to 200 feces collected for subsequent determination of parasite burden (voided feces or fecal sample via fecal loop stored in 10 % formalin), Up to 200 measurements of morphometrics (axillary girth and dorsal standard length), 90 x 2 (per animal) blubber biopsies (approx. 0.6 cm diameter, 2-3 cm in length) may be collected, 90 blood samples (up to 90 mL) may be collected,
90 swabs x 5 orifices (anal, genital, mouth, nose, eye), 200 Scats opportunistically collected on beach, Up to 90 x 2 skin plugs from flipper tagging.

There is also the possibility of conducting necropsies on any dead seals found during research activities. The type and number of samples collected during necropsies varies depending on the condition of the carcass. A necropsy protocol that highlights the potential tissues that may be collected from dead monk seals can be provided upon request, though tissues could include: samples from all major organs, skin, muscle, blood, blubber, hair, bone and other.

Collection location:
Captive Care
Potentially any of the 6 main sub-populations in the NWHI as this an opportunistic effort.

Worming Trial
Lisianski Island, Laysan Island, French Frigate Shoals

☒ Whole Organism ☒ Partial Organism

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

Samples will be analyzed on a timely basis upon return to Honolulu. For example, in 2009-2010, fecal egg presence and counts were conducted within 2 months of their return to Honolulu.

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

• General site/location for collections:
Seals could be collected from any of the major breeding populations. They will be held in shore pens or brought to the MHI to be held in pools in a captive care facility.

• 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?
They may be kept with other monk seals.

• Will organisms be released?
Yes

10. If applicable, how will the collected samples or specimens be transported out of the Monument?
All samples collected within the Monument will be transported out on the NOAA/RV OES, chartered vessel or airplane. Fecal samples will be stored in plastic-capped vials with a premeasured volume of SAF fixative, an aqueous solution of formaldehyde (10%), acetic acid and sodium acetate.

11. Describe collaborative activities to share samples, reduce duplicative sampling, or duplicative research:
Currently NOAA Fisheries is the only group researching Hawaiian monk seals eliminating duplicative research. The worming trial may share animals with foraging research to reduce human activity and the number of aniimals be utilized in monk seal research and conservation actions. However, we have several partners aiding us in the analysis of our samples and data. These include: Bishop Museum, Moss Landing Marine Lab, University of Hawaii Manoa and Hilo, UH Hawaii Institute of Marine Biology, Southwest Fisheries Science Center, Scripps Institute of Oceanography and Dalhousie University, Canada.

Data collected during this study will also be provided to the Monument to aid with their management objectives.

12a. List all specialized gear and materials to be used in this activity:
none

12b. List all Hazardous Materials you propose to take to and use within the Monument:
Up to 200 plastic-capped vials with a premeasured volume of SAF fixative, an aqueous solution of formaldehyde (10%), acetic acid and sodium acetate. Please note, technically speaking 10% formalin solutions are not considered a HAZMAT by US shipping regulations.

13. Describe any fixed installations and instrumentation proposed to be set in the Monument:
No permanent fixed installations will be set in the monument for this work.

14. Provide a time line for sample analysis, data analysis, write-up and publication of information:
Feces will be processed and logged within approximately two months of return to Honolulu. They will then be distributed to the appropriate lab for analysis. Any other samples related to captive care of seals should be analyzed within 6 months of collection depending on the workload of partner and contract laboratories. An important point to emphasize is that we do have partners in place to analyze samples and interpret resulting data.

Analysis of results of the first year of this study (August 2009 through October 2010) has been completed and a drafted manuscript is currently in internal review and set for submission for publication in a peer-reviewed journal thereafter. We expect that this additional year of tests on the efficacy of other routes of administration, dosages and medications to require at least an equal period of time, depending on results.

15. List all Applicants' publications directly related to the proposed project:
Baker, J. D.
2008. Variation in the relationship between offspring size and survival provides insight into causes of mortality in Hawaiian monk seals. Endangered Species Research 5:55-64.

Dierauf, L. A., and Gulland F. M. D.
2001. CRC Handbook of Marine Mammal Medicine. In Book CRC Handbook of Marine Mammal Medicine (Editor, ed.)^Eds.), p. 1019. CRC Press, City.

Gobush, K.S., J.D. Baker and F.M.D. Gulland.
(In preparation). The effectiveness of an anti-helmintic treatment, injectable Praziquantel, in improving the survival and condition of free-ranging juvenile Hawaiian monk seals.

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.

_____ _____

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