CAVERNS, COMPRESSED AIR AND CRUSTACEAN CONNECTIVITY: INSIGHTS INTO HAWAIIAN SPINY LOBSTER POPULATIONS

Matthew Iacchei1,2
Robert J. Toonen1

1Hawai‘i Institute of Marine Biology
University of Hawai‘i at Mānoa
Honolulu, HAWAI’I 96744 U.S.A.
iacchei@hawaii.edu

2Department of Zoology
University of Hawai‘i at Mānoa
Honolulu, HAWAI’I 96817 U.S.A.

Since the arrival of the first Polynesian voyagers to the Hawaiian archipelago spiny lobsters have proved to be valuable fisheries species in Hawai‘i. However, the combination of long-term commercial and recreational fisheries for this species, and changing environmental conditions, have led to declining catch rates since the 1950s. The Papahānaumokuākea Marine National Monument (PMNM), established in 2006, now provides an extensive harvest refuge area for lobster species in the Northwest Hawaiian Islands (NWHI). This study aims to investigate the potential for the PMNM to rejuvenate lobster populations and fisheries in the Main Hawaiian Islands by using mitochondrial DNA markers to examine how lobster populations are related throughout the Hawaiian archipelago. State and federal agencies, dive shops and recreational and commercial lobster fishermen all collaborated to obtain genetic samples across the ~2,500 km stretch of ocean spanned by the State of Hawai‘i. Over 1,249 samples from 15 islands, atolls, reefs and banks throughout the archipelago were collected prior to 2011. In the process of collecting samples for genetic analysis, we observed a significant shift in species abundance between the MHI and the NWHI. Panulirus marginatus is the most abundant spiny lobster (73%) in the NWHI catch while P. penicillatus (88%) dominates the MHI catch (Fisher’s exact test, P << 0.001). We also observed P. marginatus living at shallower depths in the NWHI where they would not be found in the NWHI. This could be due to thermal regime, habitat availability, species competition, or fishing effects.

Introduction

Both researchers and resource managers around the globe generally agree that efforts to establish effective marine protected areas require detailed information regarding connectivity among disjunct populations of species (e.g., Botsford et al., 2001; Halpern and Warner, 2003; Palumbi, 2003; Cowen et al., 2006). The size and number of connected populations and frequency of larval influxes can profoundly influence the density and persistence of local populations (MacArthur and Wilson, 1963; Stacey and Taper, 1992). This fact is particularly pertinent for benthic marine invertebrates living in isolated island chains where populations are more likely related through larval dispersal than adult movement. Unfortunately for managers this pelagic developmental period, coupled with a small size and the ability to swim against prevailing oceanographic currents, make larvae extremely difficult to track (reviewed by
Levin, 2006). The lack of knowledge of population connectivity in organisms with a pelagic larval stage (\(~85\%\) of benthic marine organisms) has caused much of the difficulty in successfully managing marine species (Carr et al., 2003). For example, the Northwestern Hawaiian Island (NWHI) lobster fishery was closed in 2000 because of increasing uncertainty in population and stock assessment models particularly with regard to spatial heterogeneity and the assumption of synchronous dynamics among bank specific populations (Botsford et al., 2002). Quantitative estimates of population connectivity are required to develop models that more accurately represent island/bank specific population dynamics.

Spiny lobsters have one of the longest larval durations of any taxa (6 months to over 1 year) making dispersal patterns in these species difficult to predict. Intuitively, species with larval durations of this length should be panmictic across broad geographic ranges (e.g., Shanks et al., 2003; Siegel et al. 2003); however, more recent meta-analysis suggests there is little to no relationship between larval duration and the degree of population structure across a species’ range (Shanks, 2009; Weersing and Toonen, 2009). Accordingly, previous Panulirus sp. genetic studies have found indications of localized recruitment despite an 8-12 month larval duration (Silberman and Walsh, 1994; Johnson and Wernham, 1999).

Hypotheses of dispersal patterns of Panulirus marginatus in Hawai’i have also been controversial and results contradictory. Pollock proposed that phyllosoma mix together in the Pacific subtropical gyre and remain there for up to four years before recruiting to Hawaiian reefs (Pollock, 1992), while MacDonald contended that larvae are retained around the archipelago for shorter time periods before settlement (MacDonald, 1986). Polovina et al. (1999) concluded that ocean currents drive phyllosoma in a southeasterly direction until they reach Necker Island, at which time they travel southwest. Previous genetic studies have failed to resolve these issues. Shaklee and Samollow (1980) used allozymes to examine the structure of P. marginatus populations across a subsection of the archipelago and suggested panmixia. Seeb et al. (1990) subsequently studied P. marginatus populations at just Maro Reef and Necker Island and found significant differences at one of seven allozyme loci but no differences at the other six. No study to date has sampled the entire archipelago nor looked at connectivity patterns in Panulirus penicillatus, a co-distributed congener that is the primary focus of recreational and commercial fisheries in the Main Hawaiian Islands.

The overall goal of this study is to determine the scales of population connectivity in the two congeneric species of spiny lobsters in the Hawaiian Archipelago, P. marginatus, and P. penicillatus, using natural genetic variation in both mitochondrial and microsatellite DNA markers. These data will enable managers to determine whether the recently established Papahānaumokuākea Marine National Monument (PMNM) has the potential to rejuvenate lobster stocks in the Hawaiian Archipelago or if additional reserves will be needed to sustain populations in the MHI. This work is ongoing and results to date are preliminary. Here we discuss the initial stages of this work, the collection of samples throughout the species range and some of the additional, previously undocumented, non-genetic results we have discovered in the sampling process.

**Methods**

**Sampling**

Lobsters were collected using three distinct methods: by hand while scuba diving or free diving, with standard Fathoms Plus® commercial lobster traps onboard both large and small research vessels, and through collaborations with commercial and recreational fishers. To cover such a broad geographical area and depth gradient for sample collection the assistance of a number of organizations was employed: two National Oceanic and Atmospheric Association (NOAA) vessels (R/V Hi‘ialakai and R/V Oscar Elton Sette), six dive shops located throughout the state of Hawai‘i, five Hawai‘i Division of Aquatic Resource (DAR) Offices, five commercial fishermen, and over 20 recreational divers/fishermen, in addition to University of Hawai‘i scientific divers. Whenever possible carapace length, sex, and a GPS coordinate of sampling locations were recorded. However, with the large diversity of sampling personnel, it was not
always possible to obtain all of this information for each specimen. All samples collected by scientists employed non-lethal sampling protocols as outlined in Skillings and Toonen (2013). In all sampling instances, a single leg or antenna segment was taken for a tissue sample and lobsters were returned to the site of capture (with the exception of commercial and recreational fishers, who retained all legal lobsters). Removal of a walking leg is a standard sampling technique for crustacean genetic surveys because these appendages typically grow back during the next molt (Mykles, 2001).

The tissue samples were taken in the field and stored in either 20% dimethyl sulfoxide salt-saturated buffer (Seutin et al., 1991; Gaither et al., 2011) or 95% ethanol at room temperature. At all locations in the Northwestern Hawaiian Islands where > 50 samples have been collected (particularly, Necker Island, Gardner Pinnacles, Maro Reef, and Laysan Island where sample sizes are >> 50) the samples were obtained from the NOAA National Marine Fisheries Service (NMFS) annual lobster tagging cruises, prior to the establishment of the Papahānaumokuākea Marine National Monument (PMNM) in 2006.

**Laboratory procedures**

Genomic DNA was extracted from each tissue sample using the DNeasy Animal Tissue kits (Qiagen Inc., Valencia, CA, USA) following the manufacturer’s protocol. For *Panulirus marginatus* a 662 bp region of mtDNA cytochrome oxidase II (COII) was amplified for each sample using the polymerase chain reaction (PCR; Saiki et al., 1988) and a standard PCR protocol (Palumbi, 1996) on a Bio-Rad iCycler Thermal Cycler (Bio-Rad Laboratories, Hercules, CA, USA). For *Panulirus penicillatus* a 460 bp region of the cytochrome oxidase I (COI) region of the mitochondrial DNA was amplified using the same procedures described above. Purified PCR products were sequenced in the forward direction with an ABI 3730XL or an ABI 3130XL capillary sequencer (Applied Biosystems, Foster City, CA). Sequences were edited, aligned, and trimmed to a uniform size using GENEIOUS PRO v. 4.8.5 (Biomatters Ltd., Auckland, New Zealand).

**Sequence analysis**

DNA sequences were edited using SEQUENCHER version 4.9b (GeneCodes Corporation). Edited sequences were evaluated using MODELTEST 3.06 (Posada and Crandall, 1998) to identify the most appropriate model of DNA evolution. Analyses are forthcoming. Briefly, we will investigate population subdivision between the MHI and the NWHI using a hierarchical analysis of molecular variance (AMOVA) (Weir and Cockerham, 1984; Excoffier et al., 1992). To assess individual population connectivity relationships we will compare pairwise population $\phi_{ST}$ values using ARLEQUIN 3.1 (Excoffier et al., 2005). ARLEQUIN will also be used to calculate nucleotide ($\pi$) and haplotype diversity ($h$) for each sampling site. Sequences will be used to create a statistical parsimony network in TCS 1.21 (Clement et al., 2000) to visually represent the relationship and distribution of haplotypes among populations. Migrate-N (Beerli, 2004) will be used to determine directionality in gene flow and quantify exchange between the MHI and the NWHI.

**Results**

**Sampling**

Prior to 2011 967 *P. marginatus* and 282 *P. penicillatus* have been collected from 15 islands/atolls throughout the Hawaiian Archipelago (Table 1). Subsequently 67 *P. marginatus* and 12 *P. penicillatus* have been collected and added to the dataset of sequences but these samples and the effort to collect them are not included in this analysis. In both the NWHI and MHI 49% of lobsters captured were female (Table 2), which was not significantly different than the expected 1:1 ratio (NWHI: $\chi^2 = 0.184$, df = 1, $P = 0.668$; MHI: $\chi^2 = 0.039$, df = 1, $P = 0.844$). Because there is a ban on taking female lobsters in the MHI the sex ratio for lobsters that can be legally harvested in the MHI (>3.25 inches carapace length (CL)) was examined. For individuals in this legal size class 45% of lobsters in the NWHI were female (Table 2), which was significantly different than the expected 1:1 ratio ($\chi^2 = 4.111$, df = 1, $P = 0.043$). In the MHI
39% of lobsters captured were female (Table 2), but this deviation was not significant due to lower sample size \( \chi^2 = 3.125, df = 1, P = 0.077 \).

Table 1. Total DNA samples collected to date from each island, atoll, and bank in Hawai‘i.

<table>
<thead>
<tr>
<th>Island Type</th>
<th>Panulirus marginatus</th>
<th>Panulirus penicillatus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Main Hawaiian Islands</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hawai‘i</td>
<td>1</td>
<td>47</td>
</tr>
<tr>
<td>Maui Nui Complex</td>
<td>17</td>
<td>84</td>
</tr>
<tr>
<td>O‘ahu</td>
<td>63</td>
<td>0</td>
</tr>
<tr>
<td>Kaua‘i</td>
<td>2</td>
<td>47</td>
</tr>
<tr>
<td>Northwest Hawaiian Islands</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nihoa Island</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Necker Island</td>
<td>161</td>
<td>5</td>
</tr>
<tr>
<td>French Frigate Shoals</td>
<td>13</td>
<td>37</td>
</tr>
<tr>
<td>Gardner Pinnacles</td>
<td>125</td>
<td>0</td>
</tr>
<tr>
<td>Maro Reef</td>
<td>232</td>
<td>5</td>
</tr>
<tr>
<td>Laysan Island</td>
<td>126</td>
<td>1</td>
</tr>
<tr>
<td>Lisianski Island</td>
<td>50</td>
<td>21</td>
</tr>
<tr>
<td>Pearl and Hermes Reef</td>
<td>67</td>
<td>30</td>
</tr>
<tr>
<td>Midway Island</td>
<td>53</td>
<td>4</td>
</tr>
<tr>
<td>Kure Island</td>
<td>57</td>
<td>1</td>
</tr>
</tbody>
</table>

Table 2. *Panulirus marginatus* and *P. penicillatus* tissue collections by sampling method, stakeholder group, sex, and size category (sub-legal versus legal-sized). Numbers listed are totals collected prior to 2011 from the Main Hawaiian Islands (MHI) and the Northwest Hawaiian Islands (NWHI).

<table>
<thead>
<tr>
<th>Catch methods</th>
<th>Panulirus marginatus</th>
<th>Panulirus penicillatus</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>MHI</td>
<td>NWHI</td>
</tr>
<tr>
<td>Snorkel/dive</td>
<td>24</td>
<td>275</td>
</tr>
<tr>
<td>Trap</td>
<td>59</td>
<td>609</td>
</tr>
<tr>
<td>Commercial Fishers</td>
<td>72</td>
<td>N/A</td>
</tr>
<tr>
<td>Recreational Fishers</td>
<td>4</td>
<td>N/A</td>
</tr>
<tr>
<td>Scientists</td>
<td>7</td>
<td>884</td>
</tr>
<tr>
<td>Males</td>
<td>3</td>
<td>276</td>
</tr>
<tr>
<td>Sub-legal males</td>
<td>0</td>
<td>72</td>
</tr>
<tr>
<td>Legal-sized males</td>
<td>3</td>
<td>204</td>
</tr>
<tr>
<td>Females</td>
<td>4</td>
<td>266</td>
</tr>
<tr>
<td>Sub-legal females</td>
<td>4</td>
<td>101</td>
</tr>
<tr>
<td>Legal-sized females</td>
<td>0</td>
<td>165</td>
</tr>
</tbody>
</table>

Specimens of at least one of the two lobster species were successfully collected while snorkeling or scuba diving at all islands/atolls in the Hawaiian Archipelago with the exception of Gardner Pinnacles and Nihoa Island. From 2006 through 2009 one of the authors (MI) was involved in dive collection trips resulting in 11,091 diver-hours searching for lobsters: 4,453 hours in the MHI, and 6,638 hours in the NWHI. A best-guess estimate places the total diver-hours spent from 2006 to 2010 searching for lobsters at close to 20,000. Diving depths ranged from 0 to 30 m, with a mean depth of 14 m (45 ft), and median of 13 m (43 ft).

In the Main Hawaiian Islands (MHI) snorkeling or scuba diving was used to collect 24 *P. marginatus*
and 175 *P. penicillatus*. In the Northwest Hawaiian Islands (NWHI) 275 *P. marginatus* and 102 *P. penicillatus* were collected while snorkelling or scuba diving (Table 2, Fig. 1). This is a highly significant shift in the species abundances across the two regions, with roughly three-fold more *P. marginatus* than *P. penicillatus* collected in the NWHI, whereas roughly seven-fold more *P. penicillatus* than *P. marginatus* were sampled in the MHI (Fisher’s exact test, \( P << 0.001 \)).

Trapping was not as widely utilized as snorkeling and scuba diving for lobsters due to difficulty in obtaining scientific collecting permits to fish with the Fathoms Plus® commercial traps. Lobsters of at least one of the two species were successfully collected with traps at six islands/atolls in the Hawaiian Archipelago and traps were fished unsuccessfully at four additional islands/atolls. Trapping depths ranged from 10 m (33 ft) to 117 m (384 ft), with a mean depth of 42 m (139 ft), and a median of 35 m (114 ft). The vast majority of lobster samples obtained through trapping were *P. marginatus* (59 samples in the MHI; 609 in the NWHI; Table 2, Fig. 1) obtained from the annual NOAA/NMFS lobster stock assessment and tagging cruises that visited the NWI each summer. Trapping proved unsuccessful for collecting *P. penicillatus* samples (3 samples in the MHI; 2 in the NWHI; Table 2, Fig. 1).

All samples collected in the Northwestern Hawaiian Islands were collected by University of Hawai’i or NOAA/NMFS biologists. In the MHI, though, both commercial and recreational fishers assisted in the sample collection. Figure 2 shows a distribution of samples collected in the MHI according to the stakeholder group that collected them.

From 2006 to 2010 261 lobster samples have been collected in the MHI: 83 *P. marginatus*, and 178 *P. penicillatus* (Table 1). Of these 59% (72 *P. marginatus*, 81 *P. penicillatus*) were collected by commercial fishers, 15% (4 *P. marginatus*, 35 *P. penicillatus*) by recreational fishers, and 26% (7 *P. marginatus*, 62 *P. penicillatus*) were collected by University of Hawai’i scientists (Table 2, Fig. 2).
Laboratory procedures

DNA was extracted from a total of 449 *P. marginatus* individuals from 14 islands/atolls and 227 *P. penicillatus* individuals from 8 islands/atolls. All of these individuals were sequenced at their respective gene (662 bp COII for *P. marginatus*; 460 bp COI for *P. penicillatus*).

Sequence analysis

Analysis of sequence data is currently underway and results will be the subject of future publications.

Discussion

Data on size, sex, and depth distributions are not usually recorded or published in marine phylogeographic or population genetic studies because these data are generally not available or relevant to the research questions being addressed. However, the collection of morphology, sex, fishery, and distribution data in addition to tissue samples for this genetic analysis has revealed interesting ecological patterns. For example, anecdotal evidence suggests that *Panulirus penicillatus* will not readily enter traps (Holthuis, 1991) but no study to date has documented this phenomenon with experimental trap catch reports. Here we report trap data (Fig. 1) that corroborates this hypothesis. Of the 673 spiny lobsters trapped in total for this study, only 5 (0.7%) were *P. penicillatus*. This result may be explained partially by the fact that the majority of lobsters (90.8%) were caught in the NWI and at deeper depths where our data suggests *P. marginatus* is the more abundant species. However, even in traps that were fished at shallower depths in the MHI, where *P. penicillatus* makes up 68.2% of the total spiny lobster catch, *P. penicillatus* only comprised 1.6% of the trap catch. These data demonstrate that while *P. marginatus* will readily enter baited traps, this method of capture is relatively unsuccessful for *P. penicillatus*. Holthuis (1991) argues that the majority of tropical spiny lobster species will not enter traps with the exception of *P. marginatus* in Hawai`i and *P. pascuensis*, which is endemic to Easter Island. In contrast, temperate lobster species support lucrative commercial trap fisheries in the North Atlantic, East Pacific, Western and Central Pacific, and South Africa (Holthuis, 1991). Physical, ecological, or behavioral drivers of these differences in trapability are unclear.
The recording of the sex of each individual lobster when possible allowed a test of sex ratio differences in the MHI and the NWHI. The NWHI have been closed to lobster fishing since 2000 (Botsford et al., 2002) but recreational and commercial lobster fishing still occur throughout the MHI. In 2006 legislation was passed banning all take of female lobsters in the MHI (previously males and females >3.25 in CL could be taken). All of the lobsters collected in the MHI for this study were obtained after the ban on female lobster take was implemented. While the sex-ratio in the MHI prior to this ban is undocumented, 49% of lobsters collected in the MHI in this study were female, with this percentage decreasing to 39% when only the lobsters of legal size (>3.25 in. CL) are considered (Table 2). This compares with 45% of lobsters >3.25 in. CL that were female in the NWHI, where there is currently no fishing (Table 2). This divergence from a 50:50 sex ratio for legal-sized lobsters in both the MHI and NWHI may be due to a number of factors including differences in species composition and growth rates, bank-specific growth-rate differences (O’Malley, 2009), or illegal and undocumented poaching.

The discrepancy in patterns seen in the MHI and NWHI could also be due to differences in sample size between legal-sized lobsters in the MHI and in the NWHI (Table 2). However, the consequences of the ban on taking of female lobsters in the MHI deserves further consideration. The continued monitoring of the ratios of female and male spiny lobsters in the MHI would provide valuable data on the effect of the State’s regulation on population dynamics of spiny lobsters here. If these management regulations continue to skew the sex ratio of lobsters so that the the majority of males are small there is the potential that female fecundity will become sperm-limited, which in turn could limit overall egg production as has been demonstrated in the congeneric P argus as well as Jasus edwardsii (MacDiarmid and Butler, 1998). This in turn would stymy any hope for stock recovery of spiny lobsters in the MHI.

One of the most interesting and unexpected results obtained from this sampling effort is the significant shift in abundance of lobster species between the MHI and the NWHI. Panulirus marginatus is the most abundant spiny lobster (73%) in the NWHI catch, while P. penicillatus dominates the MHI catch (88%). Panulirus marginatus is known to inhabit shallow depths down to 143 m, and P. penicillatus inhabits 0 to 4 m depth (Holthuis, 1991) but in the MHI it is rare to find P. marginatus shallower than 15 m. The same pattern was expected in the NWHI; however, at multiple atolls P. marginatus were commonly found in 1-3 m of water on patch reefs in calm areas inside of the atoll. Panulirus penicillatus in the MHI is commonly found in shallow depths but can be as deep as 12 m in areas of high wave action. In the NWHI P. penicillatus were much less common and where found were restricted to a much narrower depth range (5 m maximum). It is clear that P. marginatus is able to thrive at shallow depths; however, the species may have a low tolerance for turbidity and wave action.

In the NWHI the only P. marginatus found in very shallow (<3 m) waters were found in calmer areas inside of atolls where they were protected from high wave energy. However, even on the forereef where no such protection is available they could be found as shallow as 7 or 8 m. Shallow areas with low wave energy may be relatively rare in the main eight Hawaiian Islands so the species depth range has shifted deeper in the MHI. Alternatively, they may prefer cooler water temperatures and therefore need to move deeper waters in the MHI as do a number of fish species (Chave and Mundy, 1994) because surface water temperature in the MHI is warmer. It is also remotely possible that heavy fishing pressure through time has removed most of the shallow-water individuals or shifted the species distribution as a whole. Panulirus penicillatus is morphologically adapted to survive in high energy environments with legs that are both longer and greater in diameter than individuals of the same CL in other spiny lobster species (Iacchei, pers. obs.) Areas with high wave action may provide a natural harvest refuge for this species in the MHI but are more rare in the NWHI (hence the lower numbers there) although there is unlikely to be such a large discrepancy in the amount of shallow water habitat between the NWHI and the MHI. Further experiments would be required to test the hypothesis that P. marginatus outcompetes P. penicillatus where they co-occur but is excluded from high energy environments where P. penicillatus is the only
spiny lobster species present. Alternatively, the two species may have originally colonized the Hawaiian Archipelago from opposite ends of the chain and only had limited success expanding to the far opposite ends. This hypothesis will be tested to the extent possible using a phylogenetic framework with the genetic data obtained in this study. However, given the presence of both lobster species at all islands/atolls in the archipelago it is more likely that the differences in abundance and distribution are driven by ecological factors.

The genetic analyses of the *P. marginatus* and *P. penicillatus* mtDNA sequences obtained from these collected tissue samples contribute to the growing number of population genetic surveys of congeneric species in Hawai‘i (i.e., Bird et al., 2007; Craig et al., 2010; DiBattista et al., 2011; Skillings et al., 2013), leading to a broader understanding of the factors driving meta-population dynamics in the Hawaiian Archipelago. Knowledge of spiny lobster connectivity patterns will provide valuable insight into whether the protection of the PMNM will allow the rejuvenation of lobster stocks in both the NWHI and the MHI, or if further actions will be required to protect MHI lobster stocks. This added ecological component illustrates the importance and the additional insight that can be gained by recording ecological data during the collection of tissue samples for phylogeographic and population genetic studies, rather than just blindly treating each organism as “simply a bag of DNA” (M.J. Donahue, pers. com.).

**Acknowledgments**

We greatly appreciate the assistance of the following individuals in collecting specimens for this project, either through actual collection or collection facilitation: Joseph O’Malley, Bob Moffitt, John Fitzpatrick, Kona Division of Aquatic Resources, notably Brent Carman and Kosta Stamoulis, Derek Skillings, Jon Puritz, Greg Concepcion, Carl Meyer, Woody Wooderson, Illiana Baums, Jeff Eble, James Ash, George Thompson and Fathom Five Divers, Terry Buholm, Patrick Conley, Lawrie Provost and Tanya Beirne and Big Island Spearguns, Bob Carrol, Skippy Hau, Michelle Gaither, Toby Daly-Engel, Michael Stat, Meaghan Huggett, Jen Salerno, Zoltan Szabo, Jon Dale, Melanie Hutchinson, Scott Aalbers, Kacy Lafferty, Elizabeth Keenan, Molly Timmers, Daniel Wagner, Scott Godwin, Steve Karl, Kelvin Gorospe, Kevin Flanagan, Kim Tice, Miguel Castrance, Tim Clark, Josh Reese, Kim Weersing, Matt Craig, Dana Crompton, Mike Muesel, Will Love, Brian Bowen, Chris Bird, University of Hawai‘i Dive Safety Program (David Pence, Kevin Flanagan, Keoki Stender, Tina Tsubota), Scientists and crew of the NOAA ship Oscar Elton Sette (OES 07-05), Crew of the NOAA ship *R/V Hi‘ialakai*, NWHI Monument Staff, Hawaii Institute of Marine Biology office and fiscal staff, TOBO Lab members. We also thank the Papahānaumokuākea Marine National Monument, US Fish and Wildlife Services, and Hawai‘i Division of Aquatic Resources (DAR) for coordinating research activities and permitting. This work was funded in part by grants from the National Science Foundation (DEB#99-75287, OCE#04-54873, OCE#06-23678, OCE#09-29031), National Marine Sanctuaries NWHICRER-HIMB partnership (MOA-2005-008/6882), National Marine Fisheries Service, an EPA STAR Fellowship, the Watson T. Yoshimoto Foundation, the Charles H. and Margaret B. Edmondson Research Fund, the Ecology, Evolution, and Conservation Biology (EECB) NSF GK-12 fellowships (DGE02-32016 and DGE05-38550 to K.Y. Kaneshiro), and NOAA Project R/HE-6, which is sponsored by the University of Hawai‘i Sea Grant College Program, SOEST, under institutional grant no. NA09OAR4170060 from NOAA Office of Sea Grant, Department of Commerce. This is HIMB contribution No. 1567, SOEST No. 9009, and UNIHI-SEAGRANT-JC-10-21 from the University of Hawai‘i Sea Grant College Program. The views expressed herein are those of the authors and may not reflect the views of the EPA, NOAA or any of their sub-agencies.

**References**


