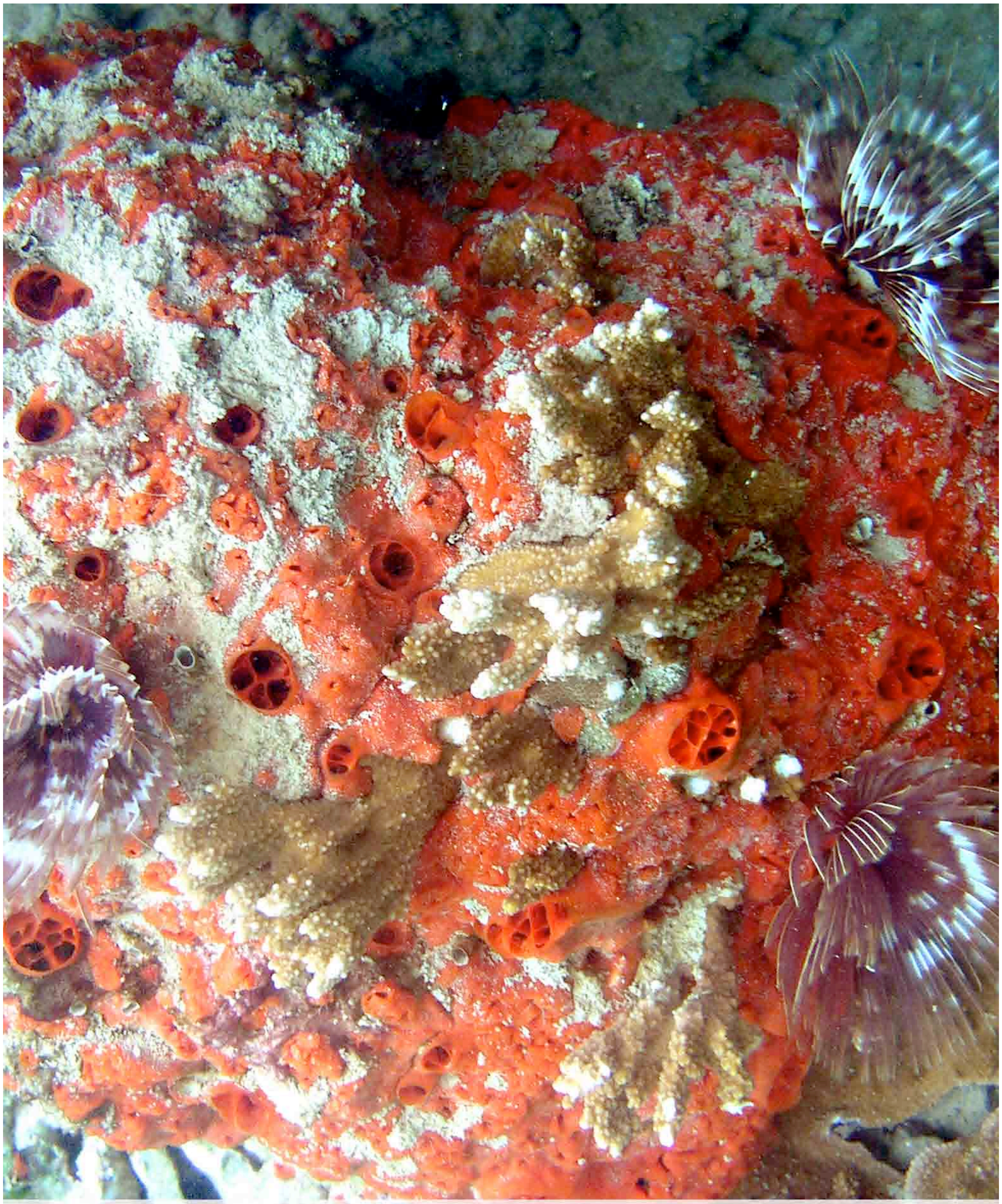

Assessment of Invasiveness of the Orange Keyhole Sponge *Mycale armata* in Kāne'ohe Bay, O'ahu, Hawai'i

Hawaii Biological Survey

Final Report, Year 2

Assessment of Invasiveness of the Orange Keyhole Sponge *Mycale armata*

in Kāne‘ohe Bay, O‘ahu, Hawai‘i.

Final Report, Year 2

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January 2007

Contribution No. 2007–002 to the Hawai‘i Biological Survey

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Executive Summary

The Orange Keyhole Sponge, *Mycale armata* Thiele, was unknown in Hawai'i prior to 1996. It was first reported in Pearl Harbor and has been reported in low abundance from a few coral reef locations near harbors, but in Kāne'ohe Bay it has become a major component of the benthic biota in the south bay in the last 5-10 years. An initial study was conducted in 2004-2005 to determine *Mycale armata's* distribution, abundance throughout the bay, its growth rates on marked permanent quadrats, and whether mechanical removal would be an effective management technique for its control (Coles and Bolick 2006). Findings in the first year from 190 manta board surveys and 19 quantitative photo-transects on 18 reefs throughout Kāne'ohe Bay indicated that the sponge had its greatest abundance in the south bay near the Hawai'i Institute of Marine Biology (HIMB) pier and Coconut Island. Despite the apparent visual dominance of this conspicuous sponge on many reefs, its maximum coverage measured on any transect in 2004-2005 was 9.2% of the bottom, with a mean of two transects at this site of 6.5%, and sponge was substantially less than coral coverage at all sites. However, measurement of changes in sponge area on ten permanent quadrats photographed quarterly throughout the year indicated a significant average increase in sponge of 13%. Attempts to mechanically remove sponge on ten other permanent quadrats was very time-consuming, requiring up to an equivalent of 22 hr m^{-2} for removal, and sponge regrew an significant average of 10% during the year following removal.

The study was continued for a second year to determine whether changes in sponge coverage and distribution in the bay could be detected, whether the first year's rates of increase in sponge cover on permanent quadrats would continue, and whether a more effective method of sponge control could be devised. Photo-transects repeated at 11 of the 19 sites from Year 1 indicated increased sponge cover at all sites with significant increases at 7 of the 11 sites, and highest sponge coverage still occurring in the vicinity of Coconut Island. The permanent control photo-quadrats remaining from the first year were re-photographed quarterly and showed a further non-significant increase of 1.7% during Year 2. Re-growth of sponge on the remaining removal quadrats averaged a non-significant increase of 6.3%. Four more photoquadrats were deployed in March 2006 and sponge surfaces on two of these were mechanically removed, followed by injection of the sponge with air delivered by a 10 cm long bone necrosis needle. This treatment resulted in mean reduction from initial values of sponge cover of up to 73% a month later. Four more quadrats were deployed in May and these were treated by air injection alone, which showed little visible effect one month later. Sponge on these quadrats were re-injected with air, and one month later showed mean reductions in sponge of 57%. Some regrowth of sponge occurred on these removal quadrats, resulting in a net average reduction of 42% below pretreatment conditions for the five of the six quadrats that remained by the end of the study.

Overall, the two-year study suggests that growth and spread of *Mycale armata* on Kāne'ohe Bay reefs and may now be slowly but steadily extending beyond its area of highest concentration in the south bay. The air injection method may provide a means for reducing the range expansion and impact of the sponge if substantial resources are directed toward controlling this highly invasive species. Before a large-scale control effort is considered, a pilot study of reducing the sponge by air injection should be conducted and results monitored to determine the effectiveness of this means of control in both the area of highest sponge abundance and at the boundary of present sponge occurrence.

Introduction

Mycale armata Thiele is an introduced sponge that is considered invasive and a potential threat to corals and reefs in Hawaiian waters (Coles et al. 2004). This species was originally described in 1903 (Thiele 1903) from a specimen collected in the Moluccas Islands in Indonesia. According to Burton (1934) this is a junior synonym for *Mycale grandis*, described by Gray (1867) from a specimen of unknown origin. However, this synonymy is considered suspect (see Hooper, et al. 1999) and, lacking further taxonomic information, *Mycala armata* is presently accepted as a valid identification for this species (J.N.A Hooper, pers. comm.)

Whatever its correct species name and type locality, it is evident that this sponge did not occur in Hawai‘i until at least the mid 20th century. Given its conspicuous characteristics and bright color, it is highly unlikely that it would have been missed by de Laubenfels (1950, 1951) on his surveys of sponges in Kāne‘ohe Bay and elsewhere in Hawai‘i. The sponge was first reported in Hawai‘i from Pearl Harbor in 1996 (Coles et al. 1997, 1999a) where it was especially prominent in the vicinity of the Hawaiian Electric thermal discharge in Pearl Harbor’s East Loch. It was first noticed in Kaneohe Bay in about 1997 (D. Gochfeld, pers. comm.) It has since been found in virtually every commercial harbor in the main Hawaiian Islands, where it can be a major component of the fouling community on harbor piers and jetties (Coles et al. 1999b, 2004). The sponge was one of the more frequently occurring introduced invertebrates on the 2003 HCRI rapid assessments of introduced species on coral reefs in the main Hawaiian islands, where it was reported at 5 of the 41 sites (Coles et al. 2004, 2006). Most of these locations were relatively close to harbors or in Kāne‘ohe Bay, where the sponge occurred at both of two sites sampled. In a more extensive Kāne‘ohe Bay study conducted in 1999-2000 (Coles et al 2002), the sponge occurred at 4 of the 24 sites sampled in the bay, and these were all in the south basin. The Guidebook of Introduced Species in Hawai‘i (Eldredge and Smith 2001) reported that the sponge appeared to be overgrowing corals in Kāne‘ohe Bay, especially the finger coral *Porites compressa* Dana and *Montipora capita* (Dana) (Frontispiece), the two dominant reef-forming coral species in Kāne‘ohe Bay.

In order to determine the spatial distribution, abundance, growth rates and competitive impacts of *Mycale armata* in Kāne‘ohe Bay, a study “Assessment of Invasiveness of the Orange Keyhole Sponge *Mycale armata* in Kāne‘ohe Bay, O‘ahu, Hawai‘i” funded by the Hawai‘i Coral Reef Initiative (HCRI) was conducted during FY2004-2005 and continued in 2005-2006. Specific tasks accomplished in the project’s first year were 1) using manta board rapid assessments, determined the spatial distribution of the sponge throughout Kāne‘ohe Bay with semi-quantitative estimates of its abundance where it occurred; 2) at selected sites based on the manta board survey results, quantitatively determined the coverage of the sponge as a percentage of the total available substrate in comparison with live coral and macroalgae; 3) using permanently marked quadrats and digital photographs, measured the coverage of both the sponge and corals quarterly to determine the degree of competition by the sponge for available habitat and if it was actively overgrowing the corals; 4) on an equal number of quadrats, measured rates of re-growth where sponge was removed in 2004 to determine whether the sponge could be effectively managed by mechanical removal; 5) quarterly measurements using buoyant weighing and estimates of sponge area from digital photographs to determine growth rates of samples of sponge collected and redeployed to the reef.

Results in 2004-2005 from 190 manta board surveys on 28 reefs and paired 25 m belt transects using photo quadrats on 19 reefs (Coles and Bolick 2006) indicated that the sponge had maximal coverage in the south-central part of the bay, in the vicinity of Coconut Island. Despite the visual dominance of this conspicuous sponge on many reefs, its maximum coverage determined on any transect was 9.2% at Reef 4, just northwest of Coconut Island, with a mean for two transects at this site of 6.5%. Sponge coverage on other reefs decreased to less than 1% on reefs at 1-3 km from this area of maximum abundance, and mean coverage for all 19 reef sites was 1.7%. Quarterly measurements of changes on sponge coverage on permanent photo quadrats indicated a mean increase of 13.1% in sponge area during one year on undisturbed control quadrats, and a mean decrease in coral cover of 16.3% on the same quadrats. By comparison, sponge coverage on quadrats where all feasible sponge had been mechanically removed at the beginning of the year showed a similar sponge mean increase of 10.2% and a mean decreased coral cover of 6.3%. These results indicate that mechanical removal would not be an effective method of controlling the sponge over the long term, especially considering that removal required an equivalent of 13-23 hours per sq. m.

These studies were continued under HCRI funding during 2006 to determine if the distribution and coverage of the sponge is spreading in the bay and if sponge growth on both control and removal quadrats has continued to increase. Also, an alternative method for removing sponge where it had already become established was tested to evaluate its management effectiveness. These findings are presented here along with recommendations for future studies and control of this invasive species.

Methods

1. Quantitative Photo Quadrat Transects

The linear photo transects method used in 2004-5 to quantitatively determine coverage of *Mycale armata* and other dominant benthic biota along transects at 19 sites on 18 reefs throughout Kāneʻohe Bay was repeated in 2006 at eleven of the sites previously surveyed (Figure 1). Digital photographs of bottom cover were taken along belt transects using an Olympus 5060 digital camera contained within an underwater housing and held on a fixed height camera stand (Figure 2), producing an image area of ca. 0.66 m^2. Twenty-five photographs were taken on each of two transects per site for a total of 33 m^2 reef surface measured per site. The start and end of each set of transects were placed to correspond with end points that had been previously recorded in 2004-5 using recorded GPS waypoints, with transect lines deployed at depths corresponding to the depths recorded for the initial 2004-2005 surveys.

Images obtained were enhanced and cropped to a consistent size of 0.66 m^2 and processed using the Coral Point Count with Excel extension (CPCe) program available from the National Coral Reef Initiative (http://www.nova.edu/ncri/research/a10.html). Each quadrat was assigned 50 random points (Figure 3) and the composition under each point was recorded, for a total of ca. 1250 points analyzed per transect or ca. 2500 points per site. These data were used to determine the percent cover of *Mycale armata*, macroalgae, corals and other invertebrates, and substratum types within the photoquadrats, which were

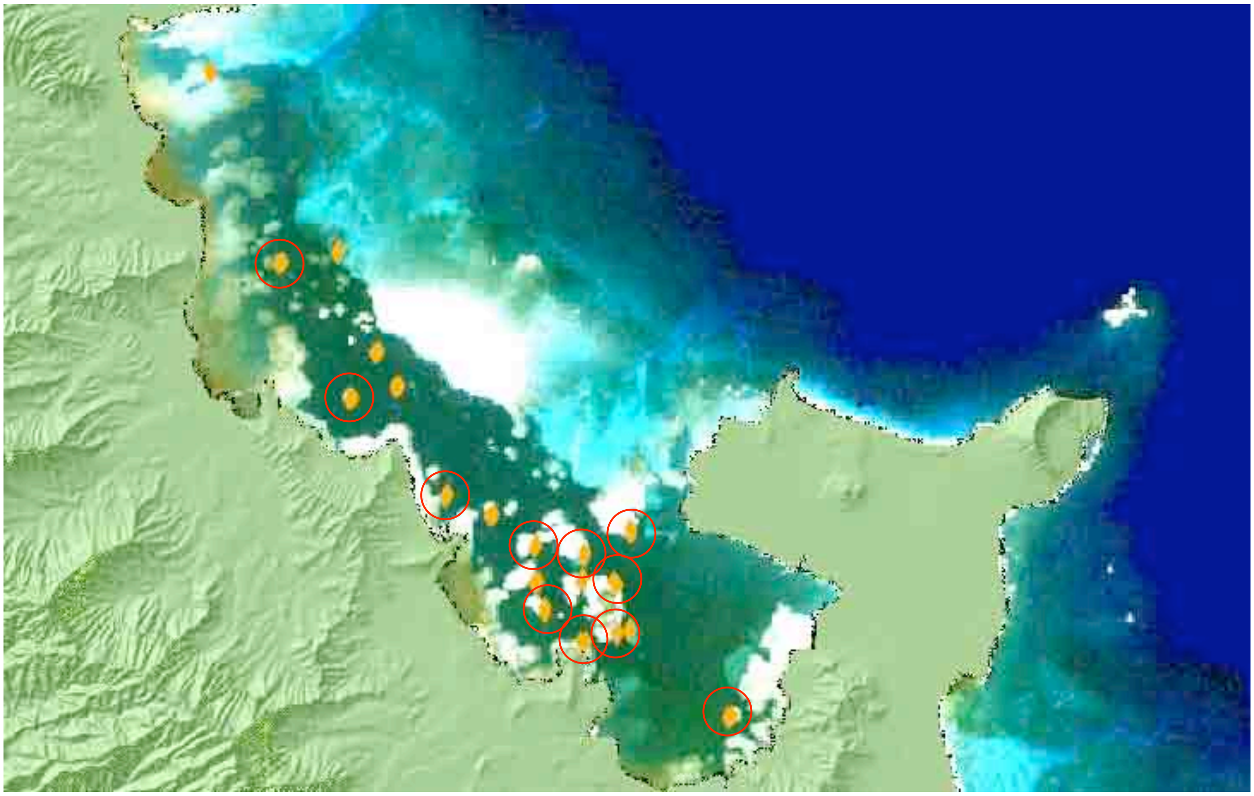

Figure 1. Locations of 19 photo quadrat transects surveyed in 2004-5(orange diamonds). The 11 sites resurveyed in 2006 are indicated by red circles.

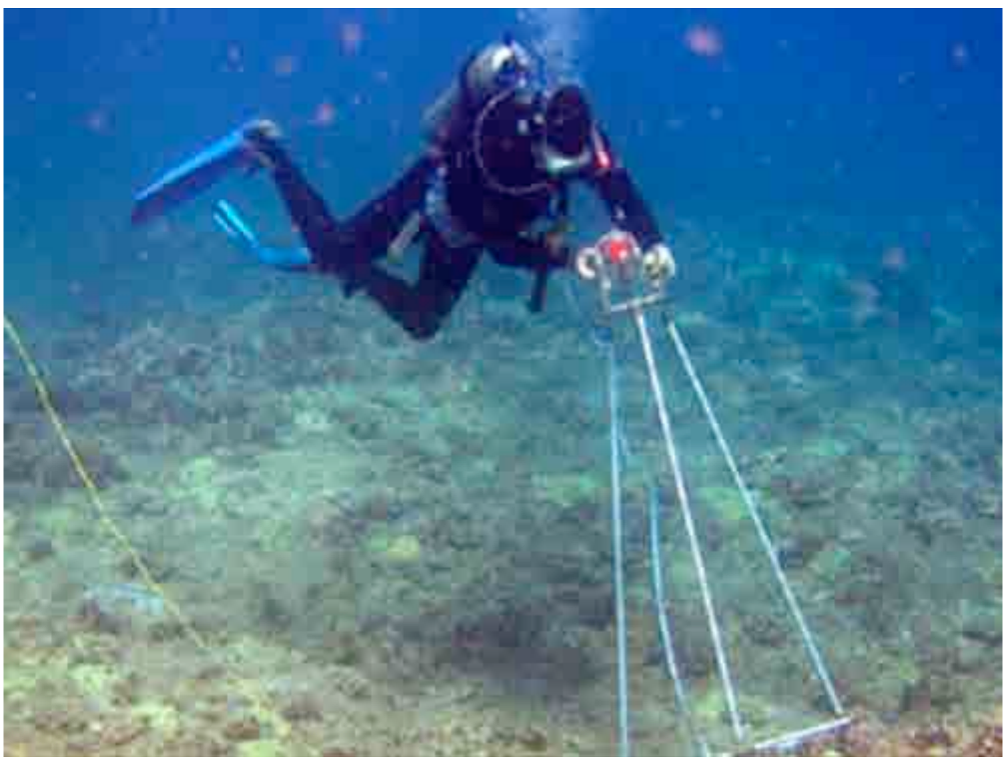

Figure 2. Diver photographing benthic cover along photo quadrat transect.

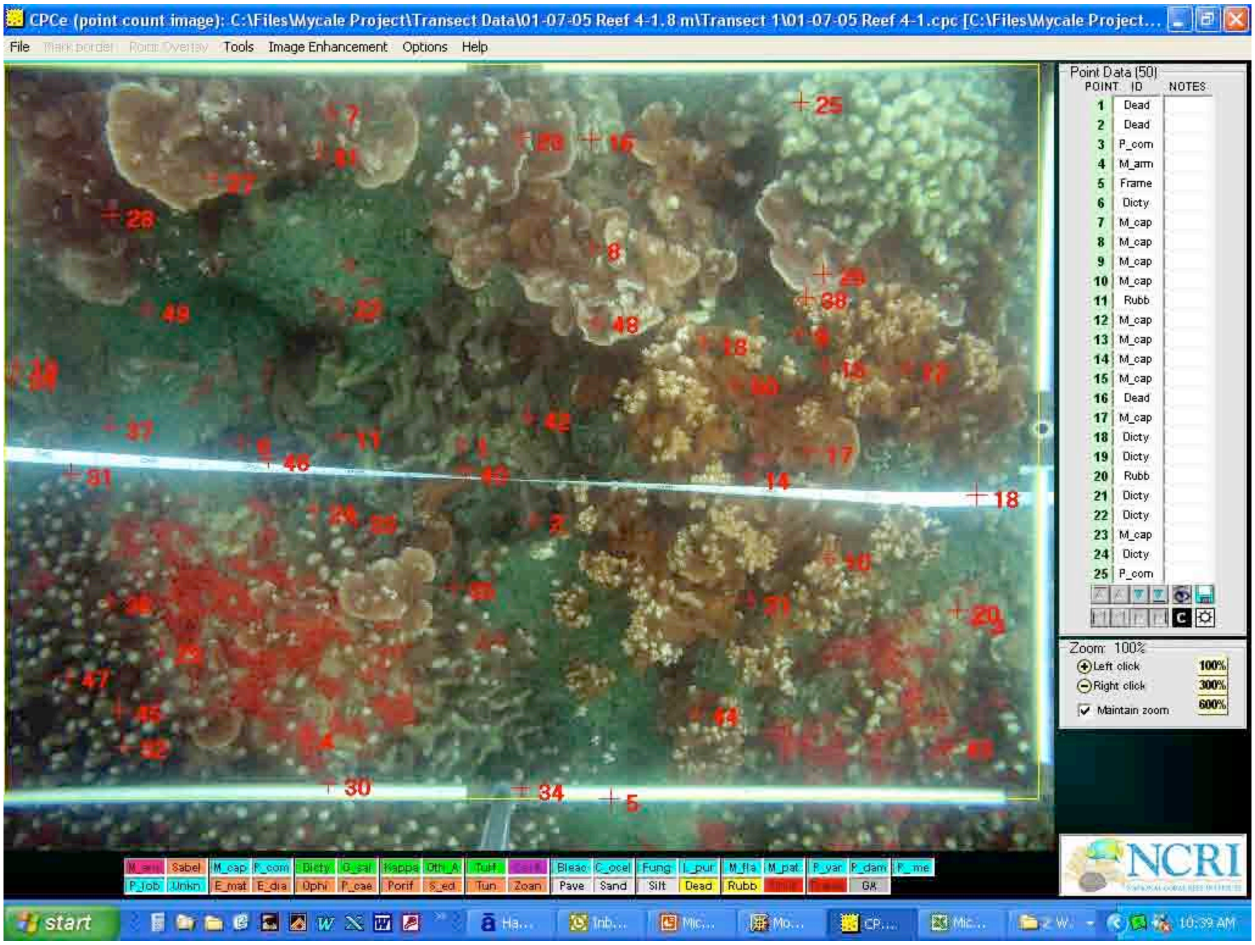

Figure 3. CPCe analysis view of a photo transect quadrat showing *Mycale armata*, *Montipora capitata*, *Porites compressa*, and *Dictyosphaeria cavernosa*.

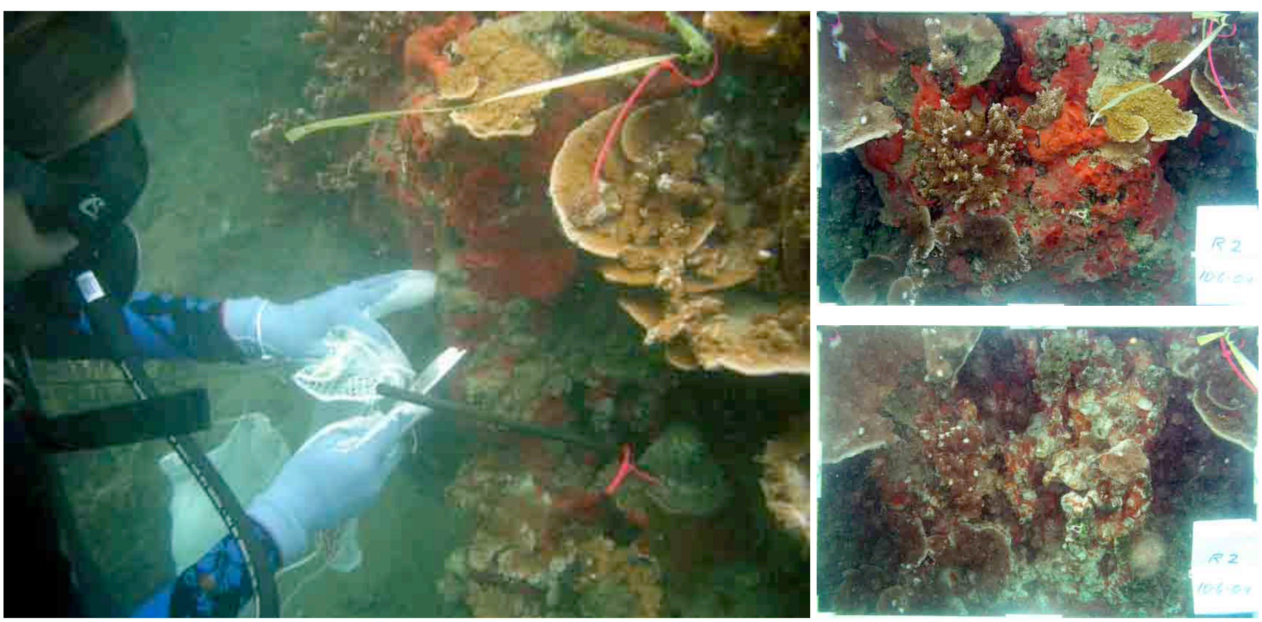

Figure 4. Sponge removal and photoquadrat showing sponge present before and immediately after removal.

averaged at each site. H. Bolick had previously analyzed all 19 phototransects sites for 2004-5 and J Marchetti analyzed the 11 phototransect sites of 2006. Since each time a photoquadrat is analyzed by this method a different set of 50 random points is projected and observers can differ in their perception of the CPCe images, especially when the imageas are of low or marginal quality, H. Bolick also analyzed five of the 11 sites for 2006 as a crosscheck for consistency of results.

2. Measurement of changes in sponge cover on control and removal quadrats

Twenty permanently marked photo quadrats were established in October 2004 on the reef slope along the southeast perimeter of Coconut Island in the vicinity of the Coconut Island 2 transects (Figure 1) to monitor sponge growth and competition with reef corals within the marked areas. The corners of 0.165 m^2 quadrats were marked with iron rebar or cable ties, enabling quarterly measurements of the same reef area for each quadrat throughout the two year study. No sponge was removed from 10 of the quadrats (Controls), while all the sponge that could be mechanically extracted from the remaining 10 quadrats (Removals) was removed (Figure 4). The amount of sponge removed averaged an equivalent of 1.85 kg dry wt per m^2 for the ten quadrats. This was time-consuming and impacted corals within some of the quadrats, requiring an equivalent of 12.6-22.7 hr m^{-2} for removal from a single quadrat and removing an equivalent 0.18-1.72 kg m^{-2} of live coral skeleton as by-catch from six of the ten removal quadrats. Both sets of quadrats were photographed soon after sponge removal and quarterly thereafter in February, May, August, and November 2005 and 2006 using a digital camera on a stand with a fixed photographic area of ca. 0.165 m^2.

After cropping to a consistent image area of 0.165 m^2, the CPCe program was used to analyze the coverage of all components within the permanent photo quadats for each quarter's determinations in a similar manner as was done for transect photo quadrats, except that 100 points evenly spaced in a grid were used for analysis of each permanent photo quadrat. This stratified approach provided greater sensitivity and probability that the same point locations on a quadrat were being sampled each quarter, resulting in more replicability and greater power for time-series analyses of changes within the permanent photo quadrats.

Of the ten control and removal quadrats originally established, nine controls and eight removals remained at the end of the first year. Those still remaining at each quarter were re-photographed, and of these, six controls and seven removals lasted until the final quarter in November 2006.

Two more control and removal quadrats were established in March 2007, and sponge was removed from these removal quadrats by a combination of first mechanical removal of the sponges surfaces, followed by injection of the sponge with compressed air from a scuba tank delivered through a 10 cm long bone necrosis needle inserted into the sponge tissue (Figure 5). Use of the needle enable delivery of the air down to the base of sponge growing on coral surfaces within the interstices of coral branches without disturbing the coral itself.

Four more removal quadrats were established on May 17, and these were injected with air without previous mechanical removal of the sponge surface to determine whether air treatment alone was sufficient to effectively eliminate sponge from a reef surface.

Figure 5. Air injection of Removal Quadrat 11 (left), appearance of quadrat immediately after air injection on March 3 (above right) and 23 days later on March 29.

Results

1. Sponge abundance on resurveyed reefs

Quantitative measurements using photoquadrat transects at 19 sites on 18 selected reefs (Figure 6) in 2004-2005 confirmed the pattern of *Mycale armata* distribution in Kaneohe Bay previously determined from manta board surveys, which indicated highest abundance in the vicinity of Coconut Island (Coles and Bolick 2006). The highest 2004-2005 value for *M. armata* determined for one transect was 9.2% at Reef 4, with a mean of 6.5% for the two transects at that reef. Mean values for the Coconut Island and HIMB dock reefs nearby ranged 4.5-5%, and *Mycale* cover decreased substantially with distance from this area to less than 1% at 1-3 km from Reef 4. Overall, *Mycale* coverage averaged only 1.7% on the 19 sites surveyed. By comparison, reef coral and macroalgae showed substantially higher coverage than *Mycale* at all sites, ranging up to 30% for total macroalgae and 89% for total coral with respective means of 7.8% and 35.3%. Coral was the dominant live bottom cover at all sites except at four reefs along the northeast side of the main ship channel in the central bay where macroalgae, with a large component of introduced and invasive species, made up the major portion of the benthic biota.

Results of resurveys of 11 of these reefs in 2006 from analysis of photo-transects by J. Marchetti showed increases in *Mycale armata* cover at all of the reefs (Figure 7), and eight of these increases were found to be significant or highly significant (Figure 8). The highest mean value determined for two transects at any reef in 2006 was 12.2% at the Coconut Island 2 reef, more than 2.5 times its previous mean value and more than twice the maximum value that had been measured for any reef (i.e. 6.5% at Reef 4i) n 2004-2005. Overall, the results showed *Mycale* to have increased substantially at many sites, with the greatest increase on the south side of Checker Reef. At that site sponge coverage determined from May 2005

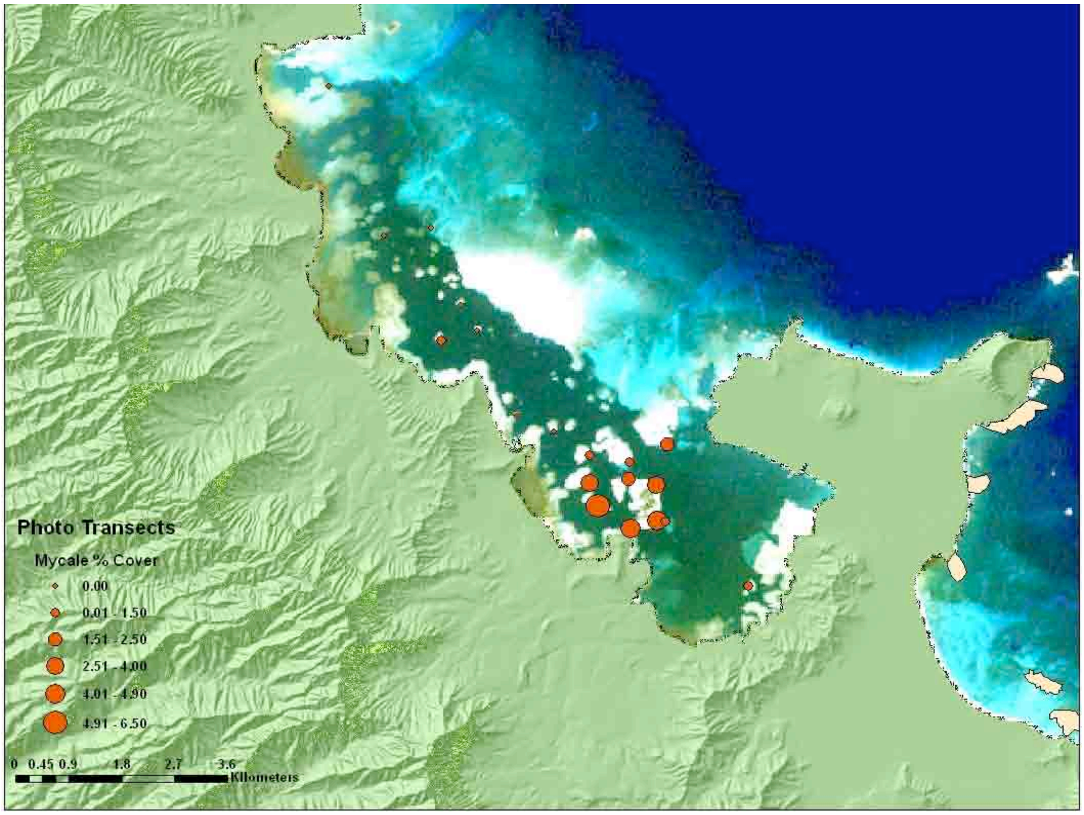

Figure 6. 2004-2005 mean coverage of *Mycale armata* determined by photo transects at 18 Kāne'ohe Bay reefs.

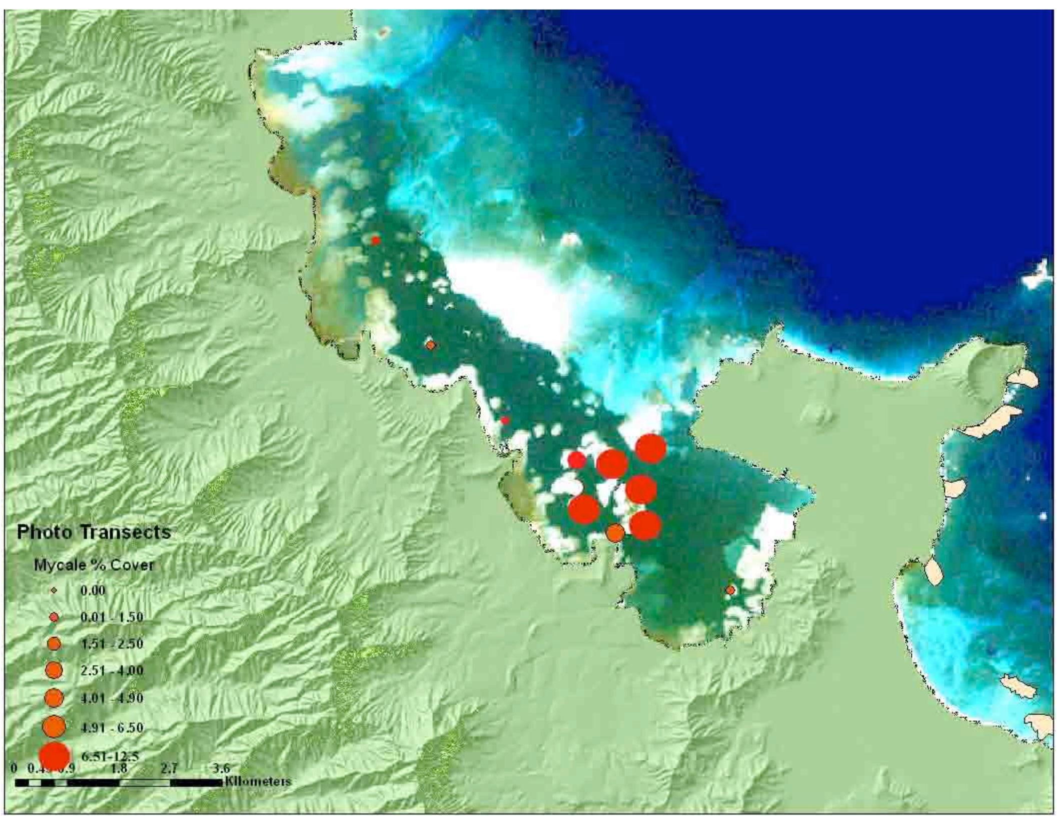

Figure 7. 2006 mean coverage of *Mycale armata* determined by photo transects at 11 reefs.

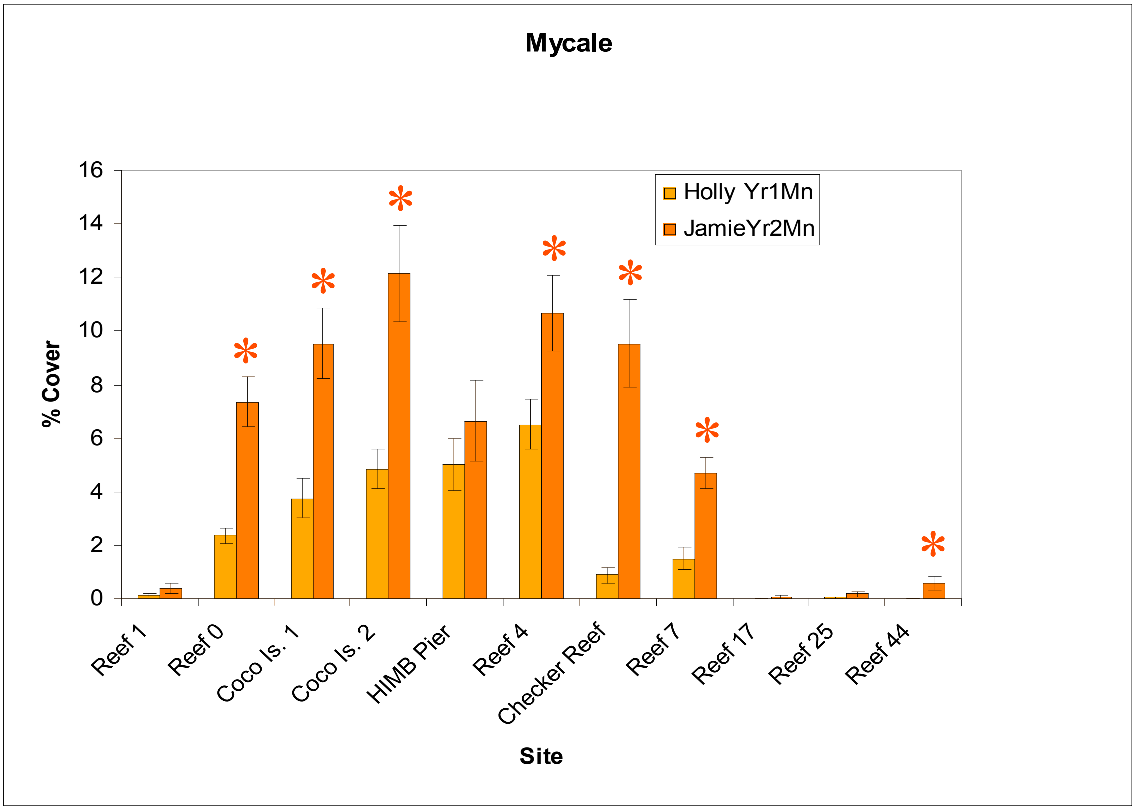

Figure 8. Comparison of means + st. errors. for *Mycale amata* cover on the 11 transects from 2004-5 that were repeated in 2006. Asteristics indicate significant or highly significant differences by 2 tailed Students t tests

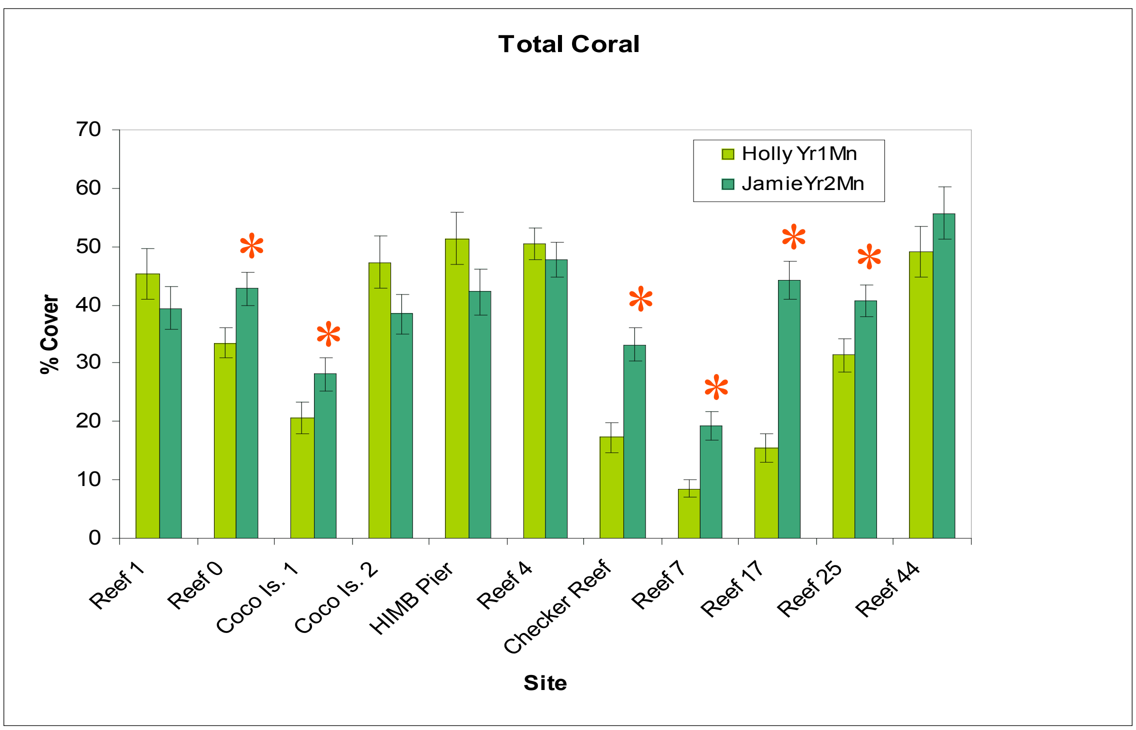

Figure 9. Comparison of Means + St. Errors for total coral cover on the 11 transects from 2004-2005 that were repeated in 2006.

photo transects by H. Bolick was <1%, and coverage determined from analysis by J. Marchetti for September 2006 photo transects was 9.5%.

Results for changes in total coverage (Figure 9) also showed increases at seven of the eleven sites, with these increases significant or highly significant at five of the sites. Coral coverage is shown to have doubled at three of the sites (Checker Reef, Reefs 7 and 17), which suggests that the placement of transects in 2006 may not have exactly coincided with transect placement during the first year's measurements. This could be one source of error between the sampling periods. The other major error source could be the use of different analysts for the two sets of measurements. To test for the latter, the results of the five sites that were analyzed by H. Bolick for 2006 transects were compared with the same transects analyzed by J. Marchetti (Figure 10). Although means determined by H. Bolick were consistently lower than found by J. Marchetti, a significant difference ($p<0.05$) was found for only one site, Reef 4, where H. Bolick's 7.0% mean was 3.6% less than the 10.6% mean determined by J. Marchetti. Comparing H. Bolicks results for 2006 for with the same five transects analyzed by her for 2004-5 (Figure 11) shows increases in *Mycale* at all sites, with a highly significant increase ($p<0.005$) at Checker Reef and an increase approaching significance ($p<0.10$) for Coconut Island 2 reef. This is a similar pattern as shown in Figure 8 for these five sites that were analyzed by J. Marchetti, except increase determined by H. Bolick for Reef 4 was not significant.

The other major potential source of error between the two sampling period was lack of consistency of transect placement. Although use of GPS coordinates and field notes assured that the transects were replicated within a few meters of their original location, slight differences in lateral placement or depth of a transect could result in major differences in results. This was tested by normalizing the *Mycale* coverage results by dividing *Mycale* values by total coral cover for each quadrat. Results (Figure 12) for analysis of 2006 quadrats by J. Marchetti indicate that *Mycale* in terms of coral cover increased at all sites similar to the pattern found for *Mycale* alone (Figure 8) and that these increases were significant at three of the 11 sites: Coconut Island 2, Checker Reef and Reef 44, where no *Mycale* had been detected on transects in 2004. Doing similar analysis for the five sites analyzed by H. Bolick for both survey periods (Figure 13) showed substantial but non-significant increases in mean *Mycale* normalized by coral cover at Coconut Island 2 and a significant increase at Checker Reef ($p<0.05$). Therefore, both raw *Mycale* coverage and *Mycale* normalized by coral suggest that the sponge was increasing cover at many sites in the south bay from 2004-5 to 2006.

2. Changes in sponge cover on permanent control and removal quadrats

Nine of the 10 control and eight of the 10 removal permanent photo quadrats remained by the time of fourth quarter coverage measurements in November 2005. Those that remained were rephotographed quarterly during 2006 and, of these, six controls and seven removals were still present at the end of the study in November 2006. Examples of the changes that occurred on control and removal quadrats through the two year period are shown in Figures 14 and 15. Although there was substantial variability in changes in cover between quarters for individual quadrats, the overall trends (Figure 16) showed consistent increases in *Mycale armata* + silt for most control and removal quadrats and decreases in total coral compared to their values during the first year. (Silt coverage was lumped with *Mycale* coverage

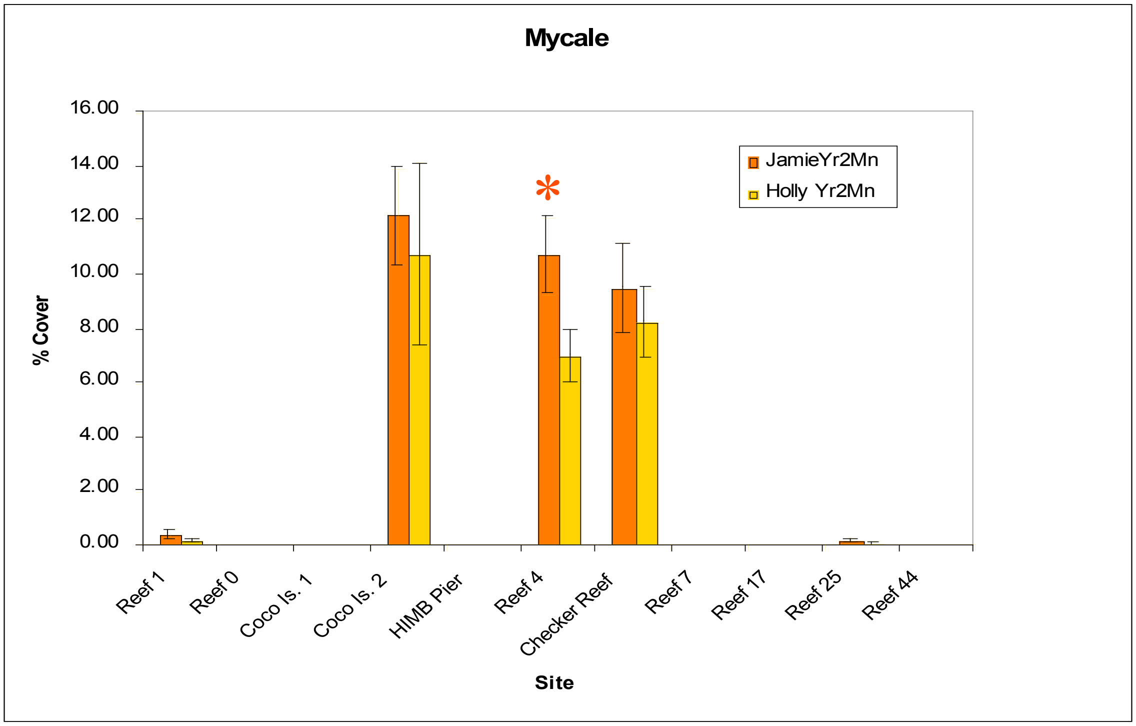

Figure 10. Comparison of Means ± St. Errors for *M. armata* cover on the 5 transects from 2006 that were analyzed by both H. Bolick and J. Marchetti.

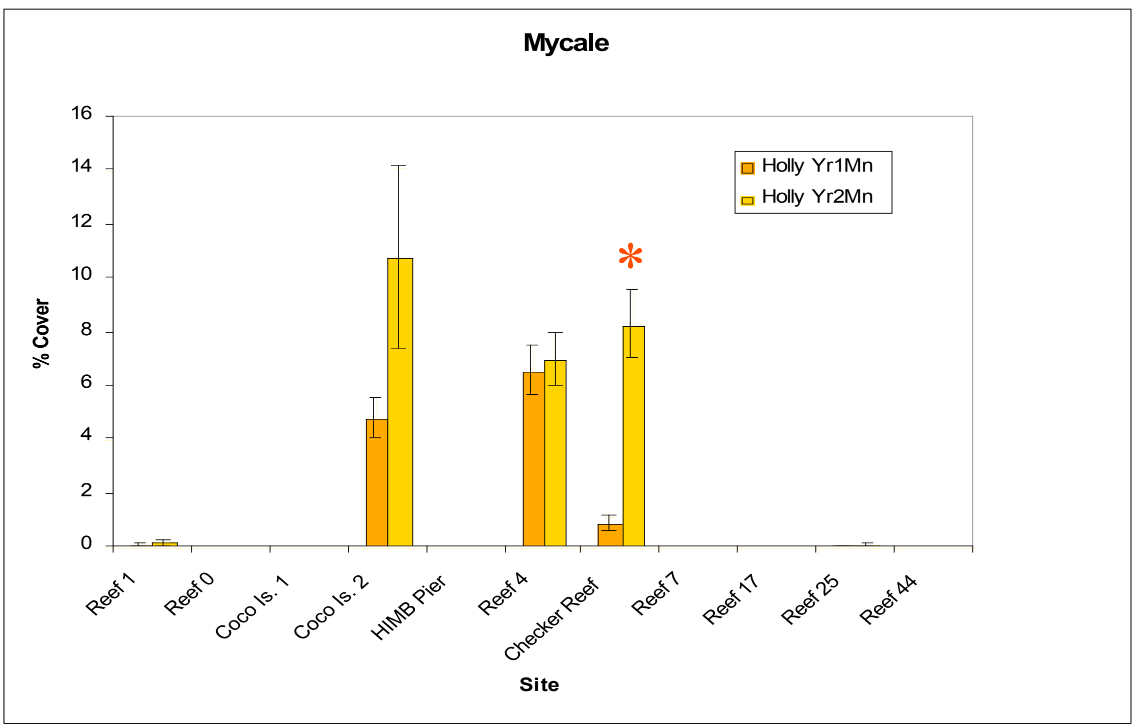

Figure 11. Comparison of Means ± St. Errors for *M. armata* cover on the 5 transects that were analyzed by H. Bolick for both sampling periods.

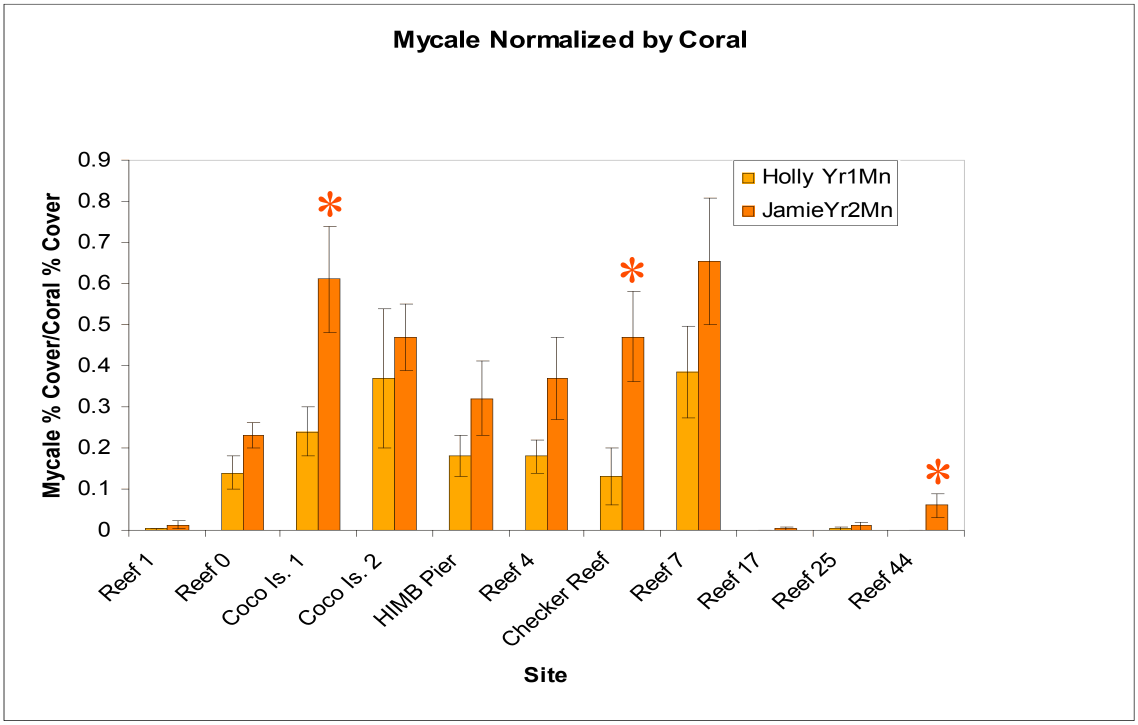

Figure 12. Comparison of Means ± St. Errors. for *Mycale amata* cover normalized by total coral cover on the 11 transects from 2004-5 that were repeated in 2006.

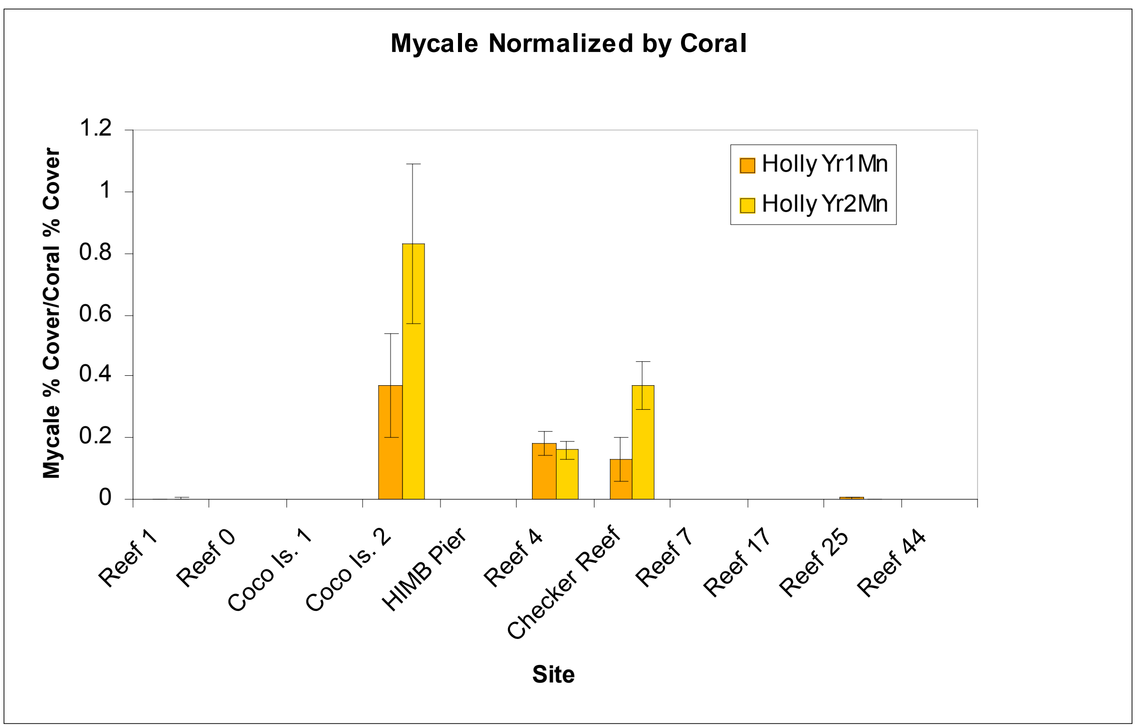

Figure 13. Comparison of Means ± St. Errors. for *Mycale amata* cover normalized by total coral cover on the 5 transects that were analyzed by H. Bolick for both sampling periods

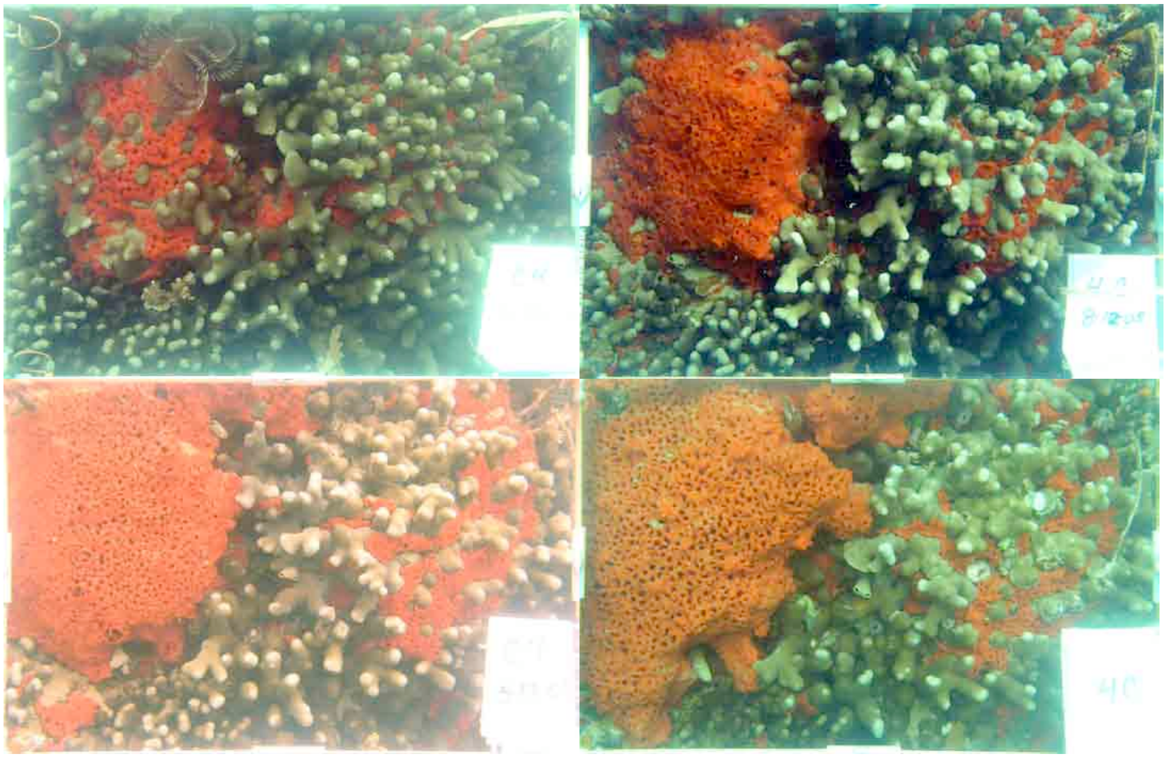

Figure 14. Semi-annual photos of *Mycale armata* on a control quadrat. a: 17Nov04; b: 11May05; c: 17May06; d: 20Nov06.

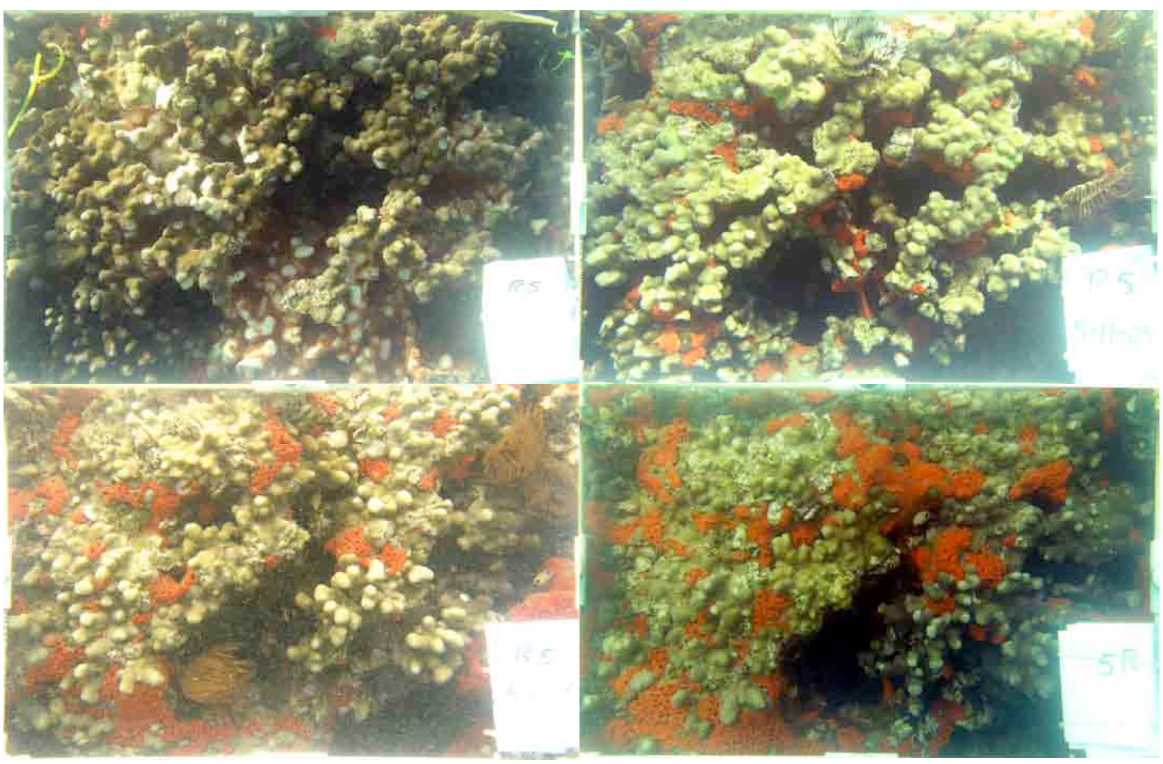

Figure 15. Semi-annual photos of *Mycale armata* on a removal quadrat. a: 22Oct04; b: 11May05; c: 17May06; d: 20Nov06

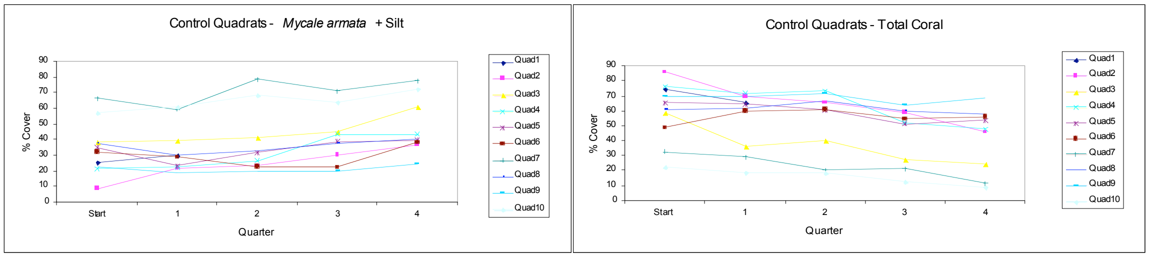

Figure 16. Time series of *Mycale armata* and coral on control quadrats Nov. 2004- Nov. 2005.

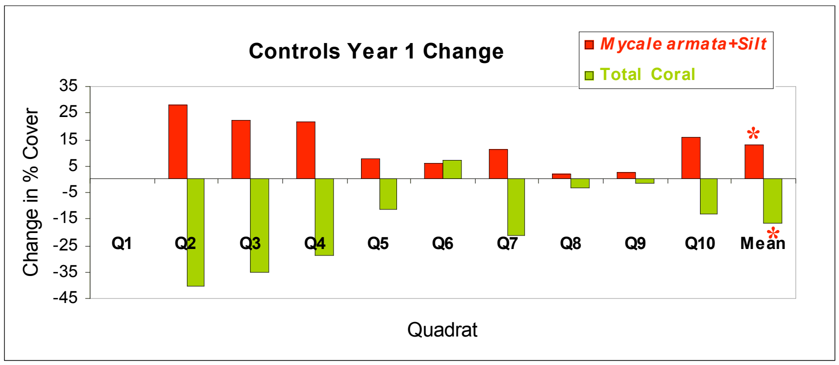

Figure 17. Year's change of *Mycale armata* and total coral on control quadrats, Nov. 2004- Nov. 2005.

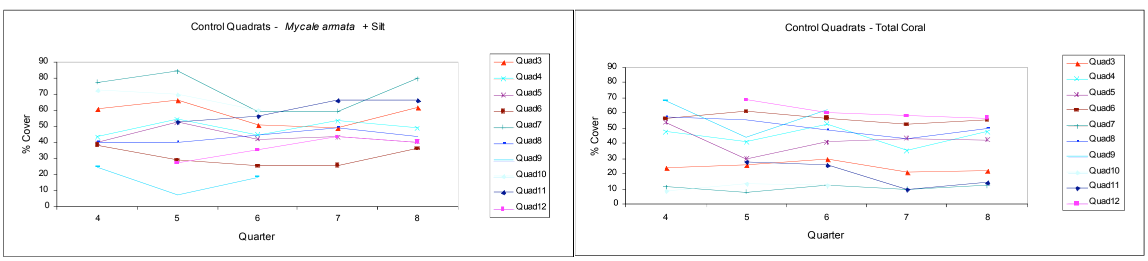

Figure 18. Time series of *Mycale armata* and coral on control quadrats Nov. 2005-Nov. 2006.

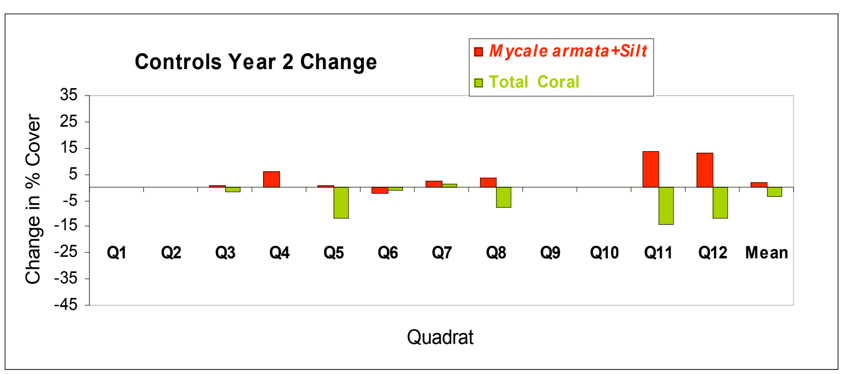

Figure 19. Year's change of *Mycale armata* and total coral on control quadrats, Nov. 2005- Nov. 2006.

because large portions of the sponge were periodically covered with silt). All nine of the control quadrats remaining by the fourth quarter (Figure 17) showed increases in *Mycale* coverage, while eight of the nine quadrats showed decreased total coral cover. These trends continued, but at a more moderate rate, during 2006 (Figures 18 and 19). The significant increases of 13% ($p<0.001$, paired t-test) in average *Mycale*+silt coverage and 16% decrease in average coral for the control quadrats during the first year (Figure 17) declined to average increase in average sponge cover of 1.7% and a non-significant decrease in coral of 3.7% during the second year (Figure 19). However, the two new controlquadrats deployed in march 2006 showed increased sponge cover averaging 13.3% and decresed coral of 12.9% by Novemver, similar to first year results for the original 10 control quadrats.

Trends similar to the above occurred for the removal quadrats during both years. Sponge coverage on quadrats that were reduced as much ca. 65% by mechanical removal in October-November 2004 (Figure 20) regrew during the first year on seven of the eight quadrats for a significant average increase ($p<0.05$, paired t-test) of 10% by November 2005 (Figure 21), with one quadrat having recovered to virtually its pre-removal value. In most cases mechanical removal of sponge from the quadrats was followed by some degree of continuous decline in coral cover, with six of the eight quadrats remaining by the fourth quarter showing reduced coral cover compared to pre-removal levels by a nonsignificant average decrease of 6.3%. During the second year, the mean sponge cover on the seven remaining removal quadrats increased a nonsignificant 6.0% and mean coral cover decreased a nonsignificant 1.4% (Figures 22 and 23).

3. New quadrats and air injection removal of sponge

The new removal quadrats that were established in March 2006 and had air injection treatment after preliminary mechanical removal of sponge surfaces remained through the end of the study. Sponge removal by this method required an equivalent of an average 3.8 hr m^{-2}, substantially less than the nearly 23 hr m^{-2} average for the mechanical removal method previously used in 2004. Four more removal quadrats were added on May 17 2006 for which air injection alone was used to remove sponge (Figure 24). Little change was noted on these quadrats when they were next seen on July 10, and the air injection was repeated. This second treatment substantially reduced sponge on all four quadrats by August 4 and three of these four remained to the end of the study in November. Sponge removal by this method of air injection without prior mechanical surface removal of sponge averaged an equivalent of ca.1.4 hr m^{-2}.

Both removal methods had highly variable results, with first quarter reductions in sponge cover below initial values ranging ca. 9-82%, followed by gradual regrowth of the sponge to end of the study (Figure 25). By November 2006 cover on the two quadrats that had received mechanical removal before air injection had net decreases of 52-78%, and two that had air injection alone showed more modest net decreases of 32-48% and one had a slight increase of 9%, for an overall mean decrease in sponge cover of 42% below initial values (Figure 26). Damage to coral by both of these methods was substantially less that by the mechanical removal method used in 2004 (Figure 17 and 19), with two of the five quadrats remaining in November 2006 showing increased coral coverage and an average increase for all five of 4.2%.

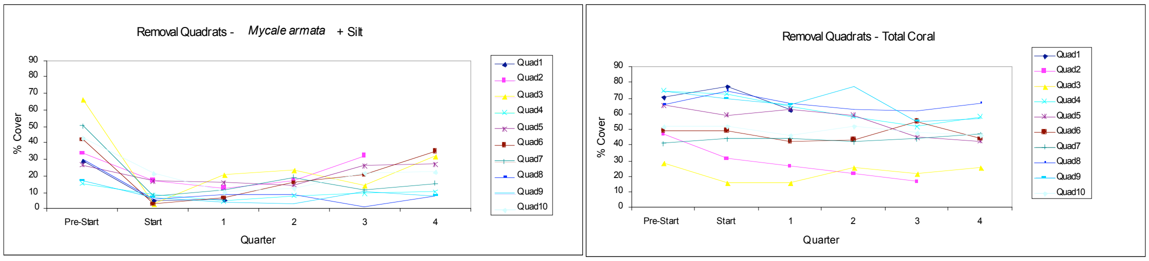

Figure 20. Time series of *Mycale armata* and coral on removal quadrats Nov. 2004-Nov. 2005.

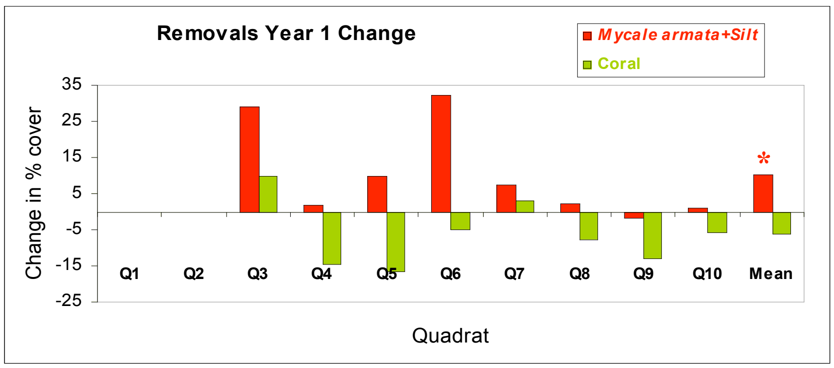

Figure 21. Year's change of *Mycale armata* and total coral on removal quadrats, Nov. 2004-Nov. 2005.

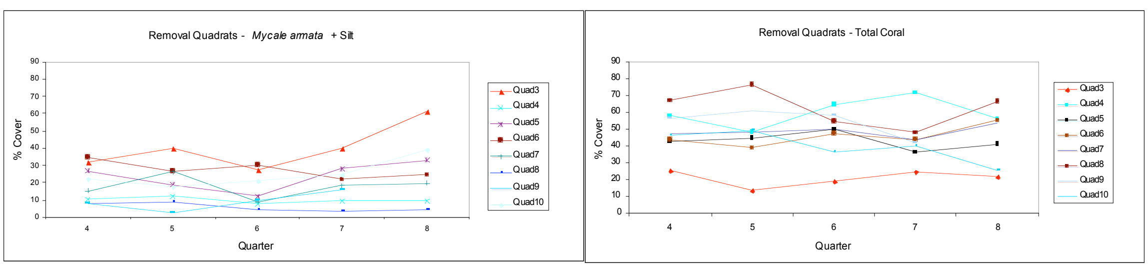

Figure 22 Time series of *Mycale armata* and coral on control and removal quadrats Nov. 2005-Nov. 2006.

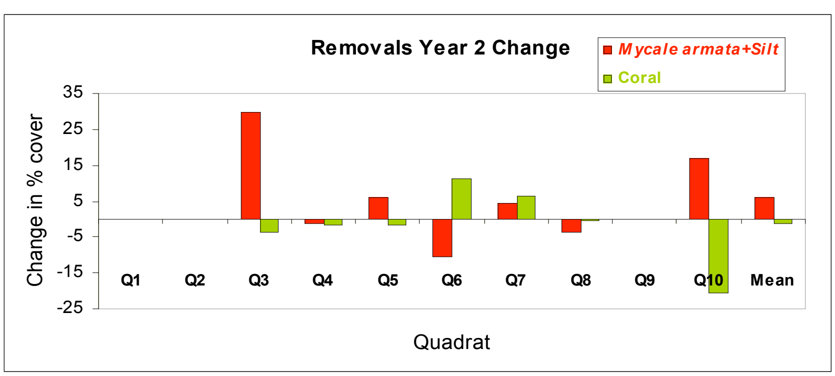

Figure 23. Year's change of *Mycale armata* and total coral on removal quadrats, Nov. 2005-Nov. 2006.

Figure 24. Sponge and coral coverage 17May, 4Aug and 20Nov on quadrat that had been air blasted in 17May.

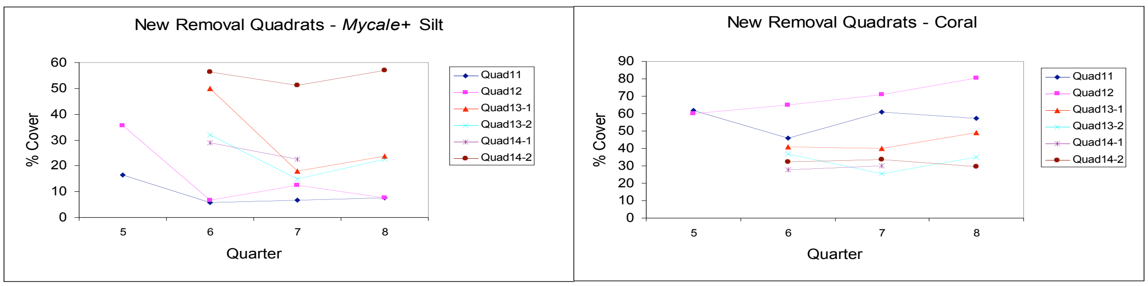

Figure 25. Time series of *Mycale armata* and coral on 2006 removal quadrats, Mar.-Nov. 2006.

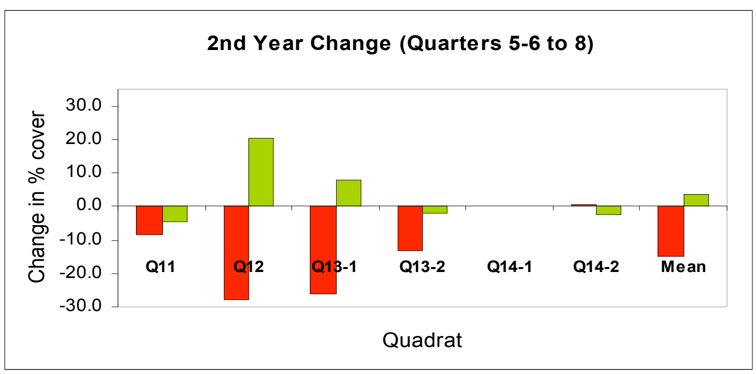

Figure 26. Change in *Mycale armata* and total coral on 2006 removal quadrats, Mar.-Nov. 2006.

4. Comparison of Analyst Consistency for Permanent Photo Quadrats

Both S. Coles and J. Marchetti analyzed a ll of the photo quadrats obtained in 2006 to determine if results were consistent between analysts and whether the methodology is therefore transferable. Results (Figure 27) show a very high consistency in analytic results for *Mycale armata*, with 96% of the variance accounted for across the full range of coverage values and a highly significant ($p<0.001$) regression coefficient with a value close to unity (1.03). The regression between analyst's results for total coral (Figure 28) was also highly significant ($p<0.001$) but with a lower variance explained of 83% and a regression coefficient of 0.89, indicating that live coral estimates by S. Coles for total coral were slightly lower than J. Marchetti for the four quarters of analysis. The agreement between analysts was lowest for both *Mycale* and total coral for the February 2006(Quarter 5) measurements when water turbidity was high and photo image quality was lowest.

Discussion

Both the highly precise photo quadrat measurements and the less precise but more extensive photo transects suggest that *Mycale armata* is increasing its coverage on reefs in south and central Kaneohe Bay and that corals on these reefs are being gradually overgrown by the sponge. The mean *Mycale* coverage on the 11 photo transects resurveyed in 2006 more than doubled in one year, from 2.3% to 5.6%, with significant increases at eight of the eleven transect sites. More importantly, the means of the ratio of sponge to coral approximately doubled, both for the eleven transects analyzed by H. Bolick for Year 1 and by J. Marchetti for Year 2 (0.148 to 0.292, Figure 11) or the five transects analyzed by H. Bolick for both years (0.137 to 0:272, Figure 12). This analysis is the most definitive evidence of increase for the transect data, since it at least partially corrects for the error inherent in not precisely replicating the location of the transect line for the second years measurements.

This indication of increasing *Mycale* coverage and declining coral coverage was supported by the results of the control and removal photo quadrats that remained through end of the second year's measurements, although rates of sponge increase and coral decrease were lower than during the first year's measurements. Sponge cover increased a mean of only 1.7% during the second year in addition to the first years 13% increase in cover on the control quadrats, and increased 6% above the first year's 10% sponge regrowth that occurred on removal quadrats, although the mean increases were not significant during the second year while they were for the first year. Comparable results for changes in total coral were a decrease of 16% in the first year followed by a further decrease of 3.7% in the second year for controls and a 6% decrease in the first year followed by another 1.4% during the second year. By contrast, a mean increase in sponge cover of 13.3% and mean decrease in coral cover of 12.9% for the two control quadrats deployed in 2006 agree well with first-year changes measured for the original ten control quadrats. These results suggest that as the sponge occupies a great area its rate of growth may slow down relative the remaining habitable space. Also, a review of quarterly measurements suggests that sponge growth is not constant but is in spurts, sometimes interspersed with periods of moderate regression. Nonetheless, the results of both photo transects and permanent photo quadrats indicate that the sponge is occupying reef space over the long term at the expense of resident native corals and may come to dominate Kāne‘ohe Bay reefs, in the vicinity of Coconut Island.

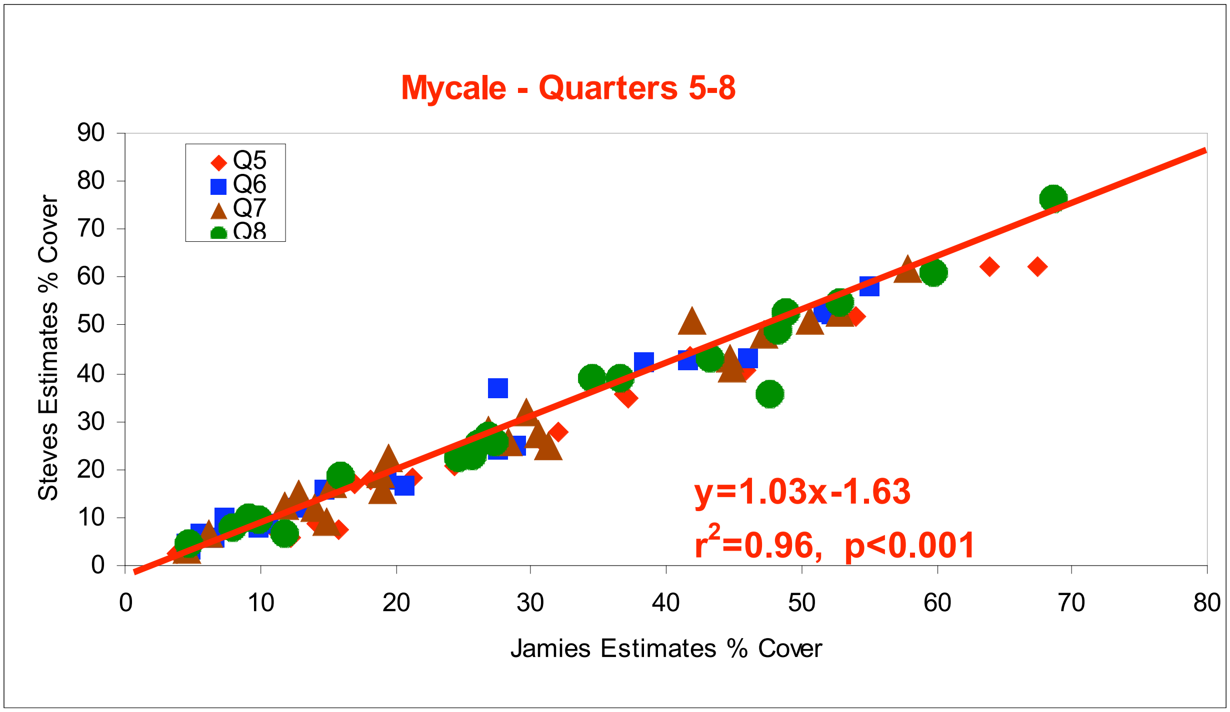

Figure 27. Comparison of results of CPCe analysis for *Mycale armata* coverage by S. Coles and J. Marchetti on 2006 permanent photo quadrats.

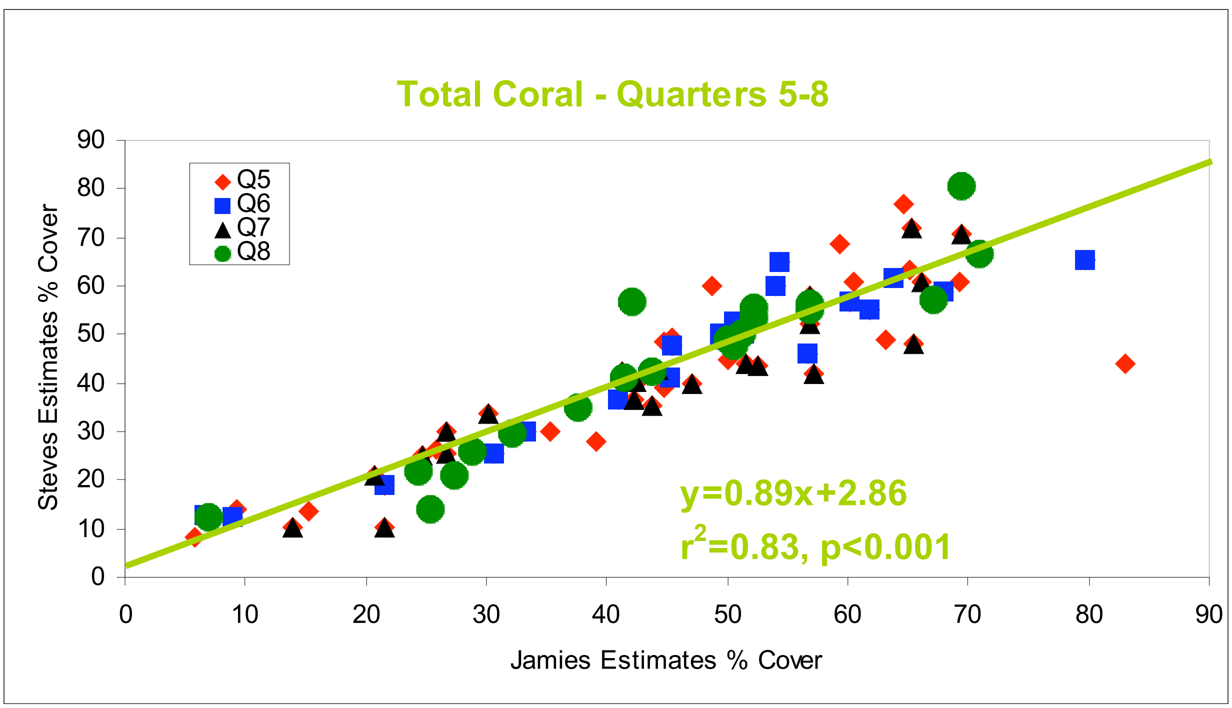

Figure 28. Comparison of results of CPCe analysis for total coral coverage by S. Coles and J. Marchetti on 2006 permanent photo quadrats.

The classic paper by Goreau and Hartmann (1966) described the ecological interaction of *Mycale laevis* (Carter) growing on the underside of corals on fore-reef slopes in Jamaica and concluded the interaction between the Jamaican *Mycale* and the coral *Montastrea annularis* to be mutually beneficial, with the sponge receiving the advantage of substratum availability free of competitors and the coral receiving increased feeding efficiency from sponge-generated water currents and protection from boring organisms. This association was manifested in a characteristic laminar form of the corals that showed prominent, regularly spaced peripheral folds at depths greater than 25 m, rather than overgrowth of the sponge surface as is the case for *Mycale armata*. Although there have been other studies of competition between corals and sponges for space on Caribbean reefs (e.g. Hill 1998, Lopez-Victoria et al. 2006) there is only one reported example of sponge-coral competition for the Pacific, *i.e. Terpios* sp. in Guam, later described as *Terpios hoshinota* (Rutzler and Muzik, 1993). This aggressive sponge grew at a rate of 23 mm mo^{-1} and rapidly spread around the west and south reefs of Guam from 1971 to 1973 to dominate shallow submerged reefs (Bryan 1973, Plucer-Rosario 1987). *Mycale armata* appears to be equally aggressive in south Kāneʻohe Bay and may eventually become dominant over native corals on Kāneʻohe Bay reefs unless an effective control method is devised.

Our results indicate that, although the sponge is still relatively restricted in the bay and occupies a relatively low percent of the available area in locations where it does occur, it is increasing on most reefs where it already occurs and may be extending its occurrence into north Kāneʻohe Bay. Not only is *Mycale armata* able to overgrow live coral completely in locations of high abundance, but it also weakens coral skeletal structure where the sponge has reached partial coverage, especially of finger coral *Porites compressa*. This rate of sponge regrowth appears to apply even when all sponge feasible has been mechanically removed.

Transect results during both survey years indicate that the sponge has its maximum abundance in South Kāneʻohe Bay near Coconut Island and decreases in coverage substantially in all directions, comprising less than 1% of the available substratum at locations more than 3 km from its epicenter in the bay. Given the nutrition requirements and ecological role of sponges as filter feeders on microscopic organic particles in Kāneʻohe Bay (Ribes et al. 2005) and the relatively higher concentrations of nutrients and particulate organic matter in south Kāneʻohe Bay compared to the mid or north bay (Smith et al. 1981, Coles and Ruddy 1995) it would be expected that high abundances of *Mycale armata* would occur in the vicinity of Coconut Island in the south bay. However, it is surprising that the sponge occurs in such low abundance at Reef 1 near the Kāneʻohe Bay Yacht Club (Figures 7-10), in the southernmost sector of the bay where organic and particulate levels in the water are maximal and there is frequent small boat traffic that could spread the sponge from fouled boat hulls. Reefs in this area are also relatively close to the Kāneʻohe Marine Base main pier and docks and boat ramps at the Marine Base, Kāneʻohe Bay Yacht Club, and Makani Kai Marinas, all of which would be likely locations from which the sponge could have been introduced to the bay. Likewise, *Mycale* cover on Reef 17 near Heʻeia public marina and boat ramps was among the lowest found during both years. Although the there is no direct evidence to confirm a hypothesis, the distribution of the sponge's occurrence and abundance suggests that it could have been introduced to Kāneʻohe Bay in the vicinity of Coconut Island or the HIMB pier.

The sponge appears to have no direct predators that would provide a natural control to its proliferation in Kāne'ohe Bay. No fishes or macroinvertebrates have been observed to feed on it, and the Hawksbill Turtle *Eretmochelys imbricata*, the only known predator of sponges (Leon and Bjorndal 2002) has not been reported in Kāne'ohe Bay and is rarely found in Hawaiian waters. However, we have found sponges with large gouges in them that appear to have resulted from recent feeding (Figure 27) near the south end of the permanent quadrats in the vicinity of the Coconut Island 2 transect site on the southeast side of Coconut Island. This location also appears to be the residential area of a green turtle (*Chelonias mydas*) with shell diameter of about 0.75 m that we have seen frequently on quarterly visits to this area, suggesting that this turtle may have at least sampled the sponge. An unidentified visitor to Coconut Island reported seeing a turtle biting a *Mycale* sponge, and caging experiments have provided some evidence for sponge feeding that might be attributed to turtles (J. Stimson, pers. coom.) If the turtle has consumed any of the sponge it has had little controlling effect, since this site (Coconut Island 2) had the highest sponge coverage (12%) determined on the second year's photo transects.

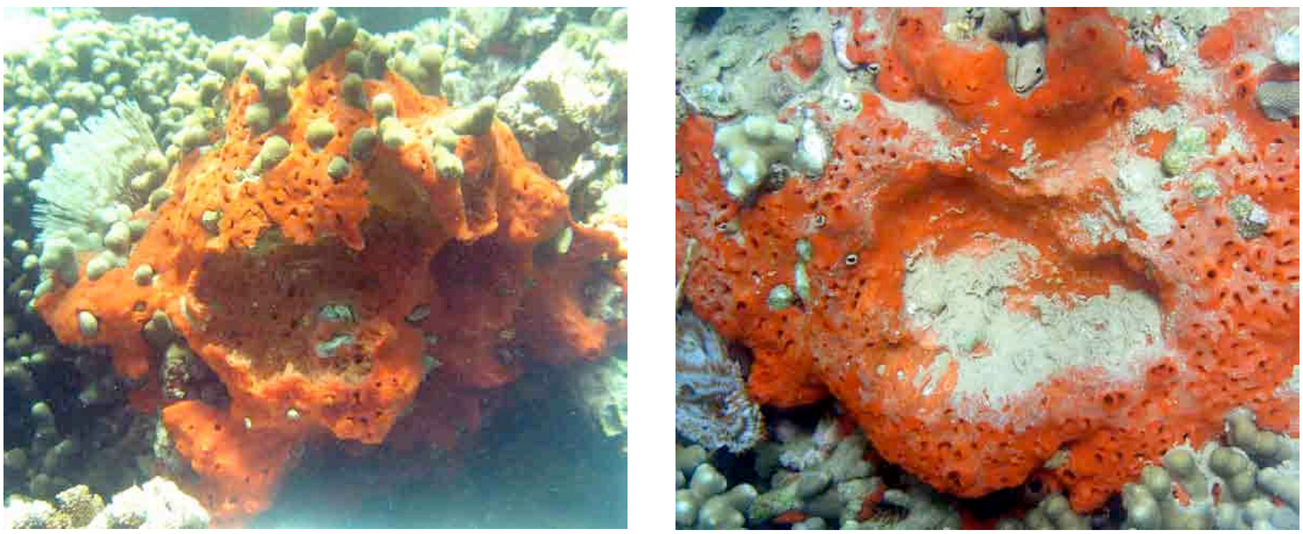

Figure 29. Cavities in sponges photographed 11May05 near permanent quadrat 10.

During the first year of this study operators of the "Supersucker" being used for mass removal of the invasive macroalga *Gracilaria salicornia* in Kāne'ohe Bay noted that the principal by-catch obtained with the algae was *Mycale armata* sponge (Eric Conklin, pers. comm., Figure 30). We noted in 2005 growth areas of this alga on the reef flat along the HIMB pier containing substantial quantities of the sponge, and that the areas of sponge sometimes remained after the algae died or was moved on by waves. Proliferation of fragments of rapidly growing *G. salicornia* that are easily moved by waves may therefore provide a mechanism by which *Mycale armata* is spread to new areas in the bay and may have produced new areas of sponge in shallow water on the reef flat along the HIMB dock causeway, over 50 m from the reef edge (Figure 30). Some of these areas of *Mycale* recruitement continue to survive in the second year, and new areas of recruitment have also been noted even further landward from the reef edge. This mechanism of transport for algal-associated *Mycale* may therefore be one means by which the sponge spreads to new areas, especially toward the leeward sides of reefs.

Figure 30. *Mycale armata* caught as bycatch with *Gracilaria salicornia* by the "Supersucker" (left), and *Mycale* growth on reef flat near HIMB dock ca. 50 m from the reef edge (right).

Management Considerations

Mechanical removal tried during the first year of the project was highly ineffective as a control of the sponge and is unfeasible in a practical sense. Divers required an equivalent of ca. 13-23 hr m^{-2} to remove as much sponge as possible from the 0.165 m^2 quadrats. This removed an equivalent of 0.6 to 5.1 kg dry wt of sponge m^{-2}, but also removed an equivalent of 0.2 to 3.5 kg dry skeletal wt. of live coral m^{-2} from 4 of the 10 quadrats as by-catch. Moreover, sponge grew back on the removal quadrats to re-occupy an average of about 10% of quadrat areas during the first year and 6% in the second, while coral cover on the same quadrats continued to decline at an average rate of about 6% in the first year and 1.4% in the second, suggesting that mechanical removal sponge imparted both initial and long-term negative impacts to corals.

The air injection technique with and without pre-removal of sponge surfaces tested during the second year of the study provides a more feasible method of reducing sponge cover in a more time and effort-efficient manner, with less collateral damage to corals. With air injection following mechanical pre-removal of sponge surfaces the time requirement was reduced to an average of 3.8 hr m^{-2}, and air-injection alone further reduced treatment time required to an average 1.4 hr m^{-2} for the first treatment, but also required an additional brief air injection one month later to produce visible sponge reduction. Similar reductions in sponge cover were achieved from both methods, resulting in decreases of -48.0 $\pm$ 11.1 % (mean $\pm$ St. Error) below initial values in the first quarter following treatment, and –42.1+12.0 % (mean + St. Error) for the five quadrats remaining at the end of the study. Both methods also produced less coral damage than mechanical removal alone. Coral cover was reduced only –6.9$\pm$8.4 % (mean $\pm$ St. Error) below initial values in the first quarter following air injection and was –8.1$\pm$8.8 % (mean $\pm$ St. Error) by the end of the study. Also, air injection did not result in breakage and release of fragments of sponge that could survive and drift to other locations where the sponge might survive and establish new colonies, as was the case with mechanical removal.

Air injection of the sponge therefore may provide a potential means of controlling growth and spread of *Mycale armata* in Kāne‘ohe Bay given enough resources. However, further testing of this method is needed to determine its long-term effectiveness and resource requirements, since sponge re-growth after air injection treatments occurred at comparable rates as followed mechanical removal. On a small scale, additional removal quadrats could be deployed and the frequency of air injection needed to permanently reduce sponge cover determined. An additional pilot study may also assist in refining this technique for its development as a new control tool for marine sponges (including as a rapid response tool to a new introduction). In order for this technique to be used on a larger-scale, the level of effort required to use this tool over a small to large spatial scale would probably be substantial. Assuming a minimum of 1 hectacre of impacted reef in Kaneohe Bay, a total of 14,000 person hours would be required to control *Mycale* utilizing this technique. This would require many qualified divers (estimated 20) with significant surface support doing repeated treatments for at least 700 hours per diver to effectively inhibit re-growth of air injected sponges and assure that all sponge locations were treated. This approach on a large-scale is therefore not practical, however, it could be tested for effectiveness for controlling small key areas of concern.

It should also be recognized that air injection or any other labor intensive method for controlling this or other invasive marine species is a palliative measure at best, and that there are almost no examples of successful control of marine invasive species after they have become established over large areas. As is the case for most introduced marine invasive species, we have little basic knowledge of the mode of introduction, life history, potential predators, or environmental conditions that favor this species that would establish a context for more effective large-scale management. Lacking this information, more research needs to be conducted on the air injection methodology in order to determine the practical use of such an approach. The alternative of no action, based on the findings of this project, may be a gradual but continuous increasing dominance of coral reefs by *Mycale armata* in Kāne‘ohe Bay.

Acknowledgments

This project was supported by funding provided by the Hawai‘i Coral Reef Initiative and the National Oceanic and Atmospheric Administration Project Numbers NA04NOS426017 and NA05NOS4261157, and by the Hawaii Invasive Species Council. We thank the staff and management of those organizations and of Bishop Museum for their continued support. Boats, scuba tanks and access to the Kāne‘ohe Bay sites were made available by the Hawai‘i Institute of Marine Biology in Year 1 and by the Hawaii Department of Natural Resources/Division of Aquatic Resoures in Year 2. We wish to specially thank HIMB Director Jo-Ann Leong and Paul Jokiel, Fenny Cox, Ku‘ulei Rogers and Fred Farrell for their tolerance of our periodic use of facilities at the HIMB "Point Lab" during Year 1. We greatly benefited from the assistance of Adrienne Romanski, graduate intern from the Dept. of Ecology, Evolution & Environmental Biology, Columbia University, in the conducting and GIS analysis of the manta board and photo quadrat surveys during the first year of the study. This internship was partially supported by the U.S. Department of Education. Special thanks to the members of the DAR Invasive Species Response Team: Cassandra Cartwright, Shawn Fujimoto, Kaylyn McCoy, and Josh Verlinde for assistance in the field, and to Steve Carroll of the Southern California Marine Aquarium Society for the initial idea of using air treatment to eliminate or control the sponge.

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