FORMAT for Annual Progress Report

A. Grant Number: NA05NOS4261157

B. Amount of Grant: $59,800

C. Project Title: **Phylogeography of Hawaiian yellow tang (Zebrasoma flavescens) with implications for management and the design of marine protected areas**

D. Grantee: Brian Bowen

E. Award Period: From: 01/01/06 To: 12/31/06

F. Period Covered by this Report: From 07/01/06 To 12/31/06

G. Summary of Progress and Expenditures to Date:

   1. Work Accomplishments: (as related to project objectives and schedule for completion)
      a. Provide a brief summary of progress, including results obtained to date, and their relationship to the general goals of the grant;

We have initiated an archipelago wide survey of Hawaiian yellow tang (*Zebrasoma flavescens*) using mitochondrial DNA (mtDNA) sequence data and microsatellite fragment analysis. The combination of highly variable nuclear markers with mtDNA analyses will allow us to resolve patterns of yellow tang larval exchange within the archipelago. The results from this project will support the management of Hawaiian yellow tang by contributing to the scientific foundations for designing marine reserves, as geography and oceanography may strongly determine the extent to which reserves can function as self-sustaining populations or enhance fisheries by supplying recruits to exploited areas.

Work/analyses completed to date:

**Sample collection**

- Sampling was conducted at multiple scales and included 553 samples collected from (a) 6 sites within the newly created Northwestern Hawaiian Islands (NWHI) Marine National Monument (1700 km), (b) 6 sites within the Main Hawaiian Islands (500 km), and (c) four sites on the largest island in the archipelago, the island of Hawaii (<100 km). Collections were also made at Johnston Atoll, which may act as a stepping stone, linking populations in the Central Pacific to Hawaii.

**mtDNA survey**

- Analysis of a 614bp stretch of the cytochrome *b* sequences demonstrates the following: (i) modest population structure across the archipelago (uncorrected Fst = 0.011; P < 0.047), (ii) no structure within the NWHI National Monument or within the main islands, and (iii) high connectivity between Hawaii and Johnston Atoll.

**Microsatellite fragment analysis**

- Through collaboration with researchers at Oregon State University we have identified 15 microsatellite loci to apply towards collected samples. The 15 loci were chosen from a candidate pool of greater than 300 loci that were developed through separate methods.
  i. 40 candidate loci were previously developed by Genetic Identification Services
ii. We isolated, cloned, and sequenced an additional 288 candidate microsatellites using the enrichment protocol developed by Glenn and Schable (Glenn, T. C. and N. A. Schable. In press [2005]. Isolating microsatellite DNA loci. Methods in Enzymology 395:202-222)

Of the 288 candidate loci we identified, we’ve narrowed our selection down to 8 loci to apply to the collected samples.

• Of the 15 selected loci, we have completed PCR optimization for 10 loci and have initiated genotyping of collected samples.

b. Provide a brief summary of work to be performed during the next year of support, if changed from the original proposal; and indication of any current problems or favorable or unusual developments; and any other significant information pertinent to the type of project support by COP, or as specified by the terms and conditions of the grant.

Funding for the project ends December 31, 2006. All work has progressed as planned with the exception of microsatellite quality control and optimization which has proceeded slower than expected. Initially we proposed to complete genotyping of samples by December 2006. Of the 15 loci we selected from the candidate pool of >300, 10 are currently being applied to collected samples and 5 are still being optimized.

The completion of the mtDNA survey of Hawaiian yellow tang provides us with an accurate estimate of the historical patterns of yellow tang larval dispersal within the Hawaiian Islands. In our preliminary mtDNA analyses we observed evidence of significant genetic partitioning indicative of reduced larval exchange among sampled sites. If subsequent analyses support our initial observations we will need to reevaluate our understanding of the basic life history of Hawaiian marine species to account for biological or physical processes that limit the free exchange of larvae within the islands.

As proposed, we intend to utilize selected microsatellite loci to assign individual recruit yellow tang to their source populations. Recruit fish collected in August will be combined with previous collections donated by Larry Basch (HCRI-RP 2002-2003). By assigning individual fish collected over multiple years to their source reefs we will be able to directly examine how recruit source may vary over space and time. These results will compliment our mtDNA based indirect estimates of larval exchange thus providing us with a robust examination of the dynamics of larval exchange among Hawaiian yellow tang populations.

2. Applications:
   a. Publications, presentations, workshops;


   b. Applications to management or research;

      Data analyses are not complete but preliminary results suggest significant genetic differentiation within Hawaiian yellow tang. Noting the existence of genetic partitions within a population is important to species management since any differentiation among sites is indicative of severely limited larval exchange. No matter whether subsequent tests continue to support our initial observation of genetic structure or rather, ultimately show all populations to be completely mixed, the data produced by this study will improve the management of Hawaiian yellow tang by providing resource managers with
a better understanding of the life history and population dynamics of this ecologically important reef fish.

c. Data and/or information products;

At the completion of the project all genetic data will be submitted to GenBank where it will be available for public access. We anticipate at least two publications in the peer-reviewed scientific literature.

d. Partnerships established with other federal, state, or local agencies, or other research institutions (other than those already described in the original proposal).

As mentioned above, we are currently collaborating with researchers at Oregon State University (Mark Hixon and his graduate student, Mark Christie) to optimize and screen microsatellite loci. Mark is currently co-PI with Brian Tissot on a project examining how individual Fish Replenishment Areas (FRAs) may differ in the selective pressures applied to incoming recruits. The synthesis of our projects will greatly improve our understanding of how physical, ecological, and behavioral factors shape yellow tang demographics; information which is critical to the successful management of Hawaii’s heavily impacted population.

3. Expenditures:
   a. Describe expenditures scheduled for this period.

   A schedule of expenditures was never established, rather, we have budgeted expenses to allow for materials, travel, and/or personal as needed throughout the term of the project.

   b. Describe actual expenditures this period.

   **Projected expenditures through 12/31/06**

<table>
<thead>
<tr>
<th>Item</th>
<th>Amount</th>
</tr>
</thead>
<tbody>
<tr>
<td>Salaries and Wages</td>
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</tr>
<tr>
<td>Material and Supplies</td>
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</tr>
<tr>
<td>Travel</td>
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</tr>
<tr>
<td>Indirect cost</td>
<td>$7,537.93</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>$45,227.57</strong></td>
</tr>
</tbody>
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   c. Explain special problems, differences between scheduled and actual expenditures, etc.

   Overall expenditures are lower than expected due to reduced travel and payroll expenses. Through collaborations with UH and visiting researchers we were able to reduce the number of outer-island trips required for sample collection resulting in lower than proposed travel expenditures. Payroll was also reduced because it was only necessary to pay a ½ year graduate assistant stipend rather than the expected full year. This was because our graduate assistant was awarded a fellowship that supported his laboratory duties for the first half of 2006. However, due to unexpected problems with microsatellite quality control and PCR optimization, expenditures on materials and supplies have been considerably greater than expected. We have submitted a budget re-allocation request.
Prepared By: Jeff Eble  
Signature of Principal Investigator  
Date 11/30/06
NOTICE

Subsequently, all NOAA COP recipients with approved grants will be asked to file a COP Annual Progress Report in the specified format. The first section of the proposed format is taken from the COP implementation plan and has some advantages in that previously-funded investigators will be familiar with the format. Consistency in reporting requirements for competitive research grant programs is desirable and this is behind COP’s efforts in proposing a standardized format. This annual report format will enable COP program staff to monitor each project supported by an award.

Public reporting burden for this collection of information is estimated to average 300 minutes per response, including time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed and completing and reviewing the collection of information.

Send comments regarding this burden estimate or any other aspects of this collection of information, including suggestions for reducing this burden, to the National Ocean Service, CSCOR/COP Office, 1315 East-West Highway, Silver Spring, MD 20910. Grant files are subject to the Freedom of Information Act (FOIA). Confidentiality will not be maintained--the information will be made available to the public. However, unpublished research results shall not be published without prior permission from the recipient.

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