

SOUTHWEST FISHERIES SCIENCE CENTER
FIRST QUARTER REPORT - FY 2005
For the Period January 1 – March 31

Submitted by: Roger Hewitt, Division Director, Fisheries Resources Division

Title of accomplishment or milestone: Molecular genetic species identification for ocean observing systems.

Current status: Ongoing. John Hyde and Eric Lynn recently returned from a billfish egg and larvae identification cruise off of Kona, Hawaii (March 11-16) on the *R/V Elton Oscar Sette*.

Background information: Ichthyoplankton samples are collected on most FRD cruises, and have been collected at least yearly since 1951. These samples provide an excellent time series and are good indicators of environmental shifts and trends in distribution and abundance of a variety of species. Traditionally, samples are collected at sea, preserved in formalin, and are brought back to the lab for sorting and identification. Identifications are generally made using a microscope and are based solely on morphology and pigmentation. This is an extremely labor intensive process, and few scientists possess the requisite expertise to identify eggs and larvae. In addition, one survey can take four to six months to process with 90% of the samples remaining unidentifiable.

Purpose of Activity: FRD is pioneering a new technique to not only improve our ability to identify ichthyoplankton samples, but to do so in real time, at sea. Samples are collected using bongo tows and the Continuous Underway Fish Egg Sampler (CUFES; Fig. 1). With an automated molecular technique eggs and larvae are processed and identified onboard the ship, within hours of collection. This new method has the potential to screen for thousands of species at once.

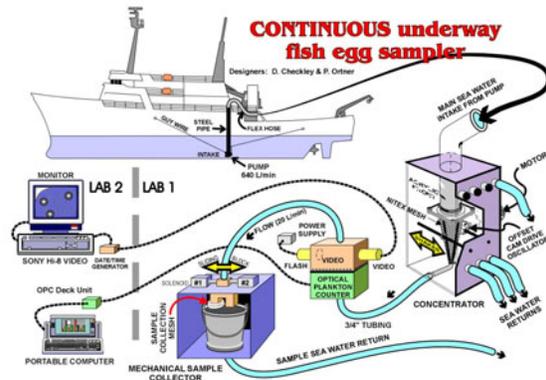


Figure 1. Schematic diagram of the Continuous Underway Fish Egg Sampler (CUFES).

Description of accomplishment and significant results: This technique was tested using 1999 CalCOFI data, which contained 4,098 *Sebastes* rockfish larvae. When identified microscopically, the larvae were sorted into 5 species, and 92% of the total samples were classified as unidentifiable. Using the new molecular genetic approach, only 2% of the larvae samples could not be identified, and those positively identified comprised 28 different species.

This new method has been field tested on several cruises off the Kona coast of Hawaii, a putative spawning ‘hot spot’ for istiophorid and xiphiid billfish. A species-specific

multiplex PCR assay was designed to amplify a single, unique size fragment of the mitochondrial cytochrome *b* gene for all 6 species of Indo-Pacific billfish, both dolphinfish, and the monospecific wahoo. Eggs and larvae were successfully identified to species within three hours of acquisition (Fig. 2). This study provided the first description of the eggs of blue marlin, shortbill spearfish, and wahoo, and is helping to determine the spatial and temporal dimension of spawning and nursery habitats of these highly valuable but poorly known species.

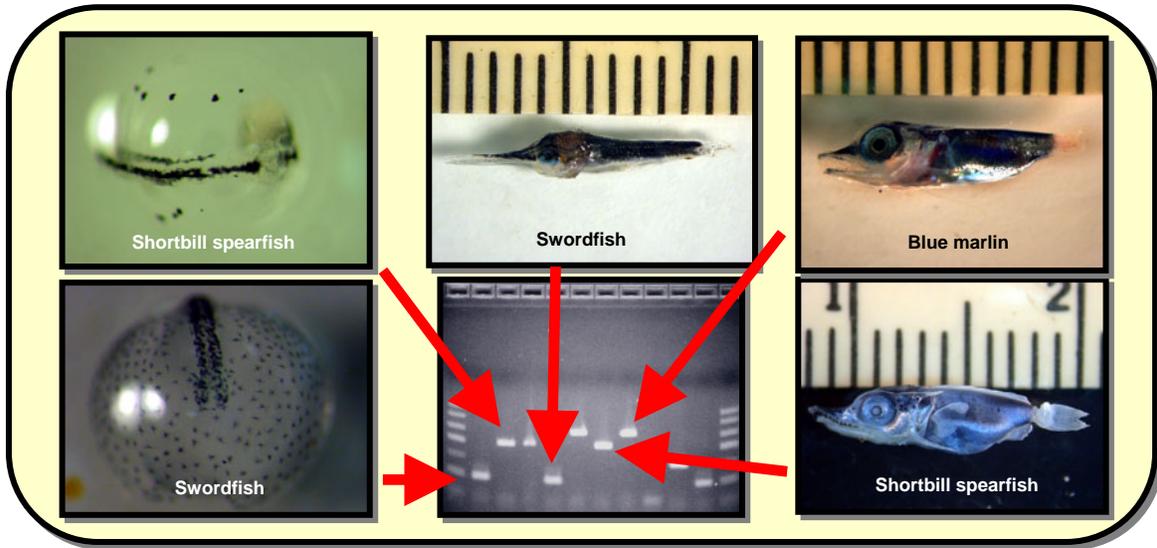


Figure 2. DNA-specific probes and optical detection identify eggs and larvae.

Significance of accomplishment: This new technique will help move management from explanation to forecasting with its ability to quickly identify large quantities of samples. This will become a necessary requirement as the ecosystem approach to management becomes more prevalent and an increased volume of PaCOOS related samples come in. Ship-board identification also allows for real-time adaptive sampling, where scientists can preferentially survey an area shown to contain the desired species. This ability to better identify eggs and larvae also gives us an improved understanding of early life history of over-fished stocks. In addition, this technique can be used for forensic identification and will make possible egg production estimates for previously unidentifiable species.

Problems: None.

Contact: John Hyde 546-7086; Russ Vetter 546-7125.