HSWRI Aquaculture Program Research Report
**** December 2013 & January 2014 ****

A Comprehensive Evaluation of Egg Quality in Three Species of Marine Fish

HSWRI recently launched a three year project funded by the Western Regional Aquaculture Center (WRAC) to study egg quality in three different marine fishes cultured for food. The species include the pelagic California yellowtail (*Seriola lalandi*; YT; Figure 1); a migratory deep-sea whitefish, the sablefish (*Anoplopoma fimbria*; SF; Figure 2); and a benthic flatfish, the California halibut (*Paralichthys californicus*; CH). Collaborating with HSWRI on the project are two NOAA Laboratories – the NW Fisheries Science Center and SW Fisheries Science Center, the University of Idaho, and California Sea Grant. Previous research by HSWRI and others indicates that egg quality can have a significant effect on larval and juvenile quality and rearing success. The egg-related factors of interest include fecundity, egg maturity, fertilization rate, egg size, lipid and fatty acid quantity and composition, and cleavage morphology to name a few. These egg quality traits may themselves be influenced by many factors, including diet, genetics, maternal effects (size, age, condition, etc.), pathogens, spawning time, spawning method, and even environmental parameters like temperature and water quality.

Figure 1. HSWRI researchers collect gametes from a young California yellowtail to determine its sex.

Figure 2. NOAA researcher Ken Massey weighs a female sablefish that they just spawned.

Egg quality directly affects the reproductive success of individuals in a breeding population, as well as the fitness of their offspring. This will, in turn, have a direct economic impact on an aquaculture program as a consequence of product quality. However, there are a myriad of ways in which the egg characteristics themselves are influenced and through which the resulting egg quality can be measured. Consequently, we still do not always understand across species how to predicatively determine which eggs or spawns are high quality and optimal for juvenile propagation or whether there are particular metrics that are more generally predictive than others. This remains a critical gap in our understanding of how to reliably and consistently culture healthy juvenile fish. We intend to address this lack of knowledge through the research in this three year project, thereby advancing aquaculture development for marine finfish along the U.S. West Coast and elsewhere in the United States.

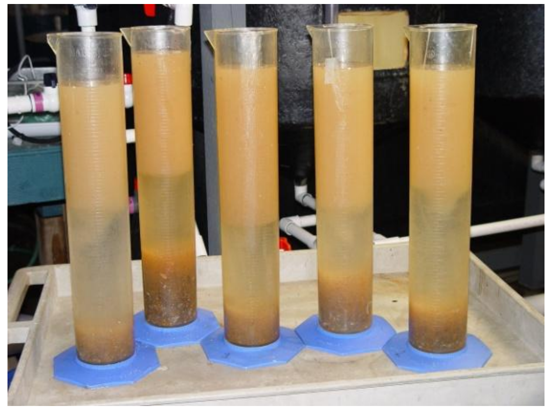

Figure 3. Eggs are enumerated volumetrically in graduated cylinders. Note the floating fraction of healthy viable eggs at the top and the non viable eggs at the bottom of each cylinder.

Ocean Acidification

Ocean acidification, caused by elevated atmospheric carbon dioxide (CO_2) being assimilate into coastal waters, is documented to result from anthropogenic processes (e.g. burning of fossil fuels). It is estimated that the increasing concentrations of atmospheric CO_2 will result in higher oceanic levels of dissolved CO_2, with subsequent lowering of seawater pH (this is often referred to as "ocean acidification"). In addition to coastal fluctuations in CO_2 and pH, conditions of low pH are common in aquaculture settings where fish are raised at high densities in tanks. Elevated CO_2 concentrations are of concern with regard to finfish health, growth and development. Some studies have documented effects of ocean acidification on invertebrates and bivalves, but limited research has been performed on finfish.

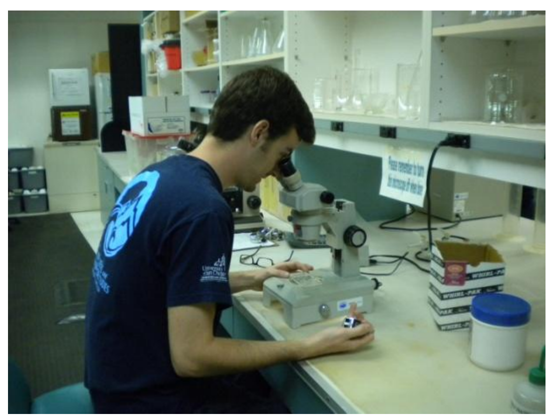

Figure 4. USD graduate student Anthony Basilio examines a white seabass larva under the microscope.

Hubbs-SeaWorld Research Institute has recently teamed up with researchers at USD to study the effects of elevated CO_2 concentrations in the seawater environment and its effects on fish eggs and larvae. The USD research team includes Dr. Nathalie Reyns, Dr. Steven Searcy and graduate student Anthony Basilio (Figure 4). This study uses white seabass as a model species to examine the behavioral effects of ocean acidification on fish larvae; specifically documenting behavior and the larvae's ability to capture prey successfully. After having learned how to raise and maintain white seabass larvae in a biosecure environment, Anthony is now beginning to mimic ocean acidification conditions by infusing CO_2 at various levels into the water of larval tanks. Anthony will then record larval behaviors during feeding beginning at 3 days post hatch, and thereafter at regular age intervals. Feeding success, weight and length of larvae will be documented throughout the culture process to assess effects of elevated CO_2 on larval growth and development. This research will lead to a better understanding of how fish larvae are affected by increased atmospheric and oceanic CO_2 concentrations and the potential repercussions of future ocean acidification on marine fish populations.

Clearing and Staining as a Tool for Assessing Malformations in Fish

Physical abnormalities, also known as malformations, can commonly affect cultured finfish and are widely varied and typically occur in the embryonic or early juvenile stages of development. Many variables are known to cause malformations including nutrition, handling, and water temperature, currents and quality. The majority of malformations in juvenile fish appear to impact bone development, particularly those comprising the head region. HSWRI recently formalized a Quality Assessment and Control Program (QA/QC) that hatchery workers use to consistently identify and remove malformed fish so that only the highest quality fish are released into the ocean and their post-release survival is not impaired. This procedural document is being coupled with complementary research designed to minimize malformations among all species cultured at HSWRI.

Several methods are available to examine the bones in fish (e.g. radiographs, computed tomography); however their successful application varies with fish size and degree of bone development. One method successfully used on small fish (i.e., <100g) is to clear the tissue and muscles and stain the cartilage and bones (Figure 5). Commonly referred to as *clearing and staining*, this method immerses the fish in multiple chemical baths during a month long process that yields a "see-through" fish with red and blue colored cartilage and bone. There are many variations on this method and each species of fish reacts differently to the chemical process so it becomes somewhat of an art form to achieve a good specimen.

Figure 5. A 50 day old white seabass with a dorsal head indent malformation in the process of clearing and staining.

HSWRI research assistant Genevieve Rich has been working through this process with 50 day old white seabass to determine what works best for this species. As the malformation assessment continues at HSWRI, viewing these cleared and stained specimens will assist in our identification of the bones that are malformed. Understanding when and where these malformations occur during the culture process is an important element in helping to control or eliminate the factors that may cause them.

Acknowledgements

This document reports on aquaculture research projects supported by numerous grants, contracts and private contributions. It also represents the hard work of many dedicated staff and volunteers throughout southern California, as well as collaborators around the country. This information was contributed by HSWRI staff and compiled by Senior Research Scientist and HSWRI Aquaculture Program Director Mark Drawbridge.

The aquaculture research program has been active for more than 35 years at HSWRI. The primary objective of this program is to evaluate the feasibility of culturing marine organisms to replenish ocean resources through stocking, and to supply consumers with a direct source of high quality seafood through aquatic farming. Please direct any questions to Mark Drawbridge at mdrawbridge@hswri.org.

Aquaculture research at HSWRI is currently supported by these major contributors:

- Avalon Tuna Club Foundation
- Cabrillo Power/NRG
- California Sea Grant
- Chevron Corporation
- NOAA's Saltonstall-Kennedy Program
- San Diego County Fish and Wildlife Advisory Commission
- Santa Monica Seafood
- SDG&E Environmental Champions
- SeaWorld Parks and Entertainment
- SeaWorld San Diego
- Soy Aquaculture Alliance
- The California Department of Fish and Wildlife's Ocean Resources Enhancement and Hatchery Program
- The Catalina Seabass Fund
- The Fletcher Foundation
- The Shedd Family
- The U.S. Fish and Wildlife Service's Sport Fish Restoration Account
- United Soybean Board

- USDA National Institute of Food and Agriculture
- Western Regional Aquaculture Center (WRAC)

Hubbs-SeaWorld Research Institute is a 501(c)(3) non-profit charity. If you would like to become a financial supporter of the Institute's aquaculture research, please contact HSWRI's Development Director, Eileen Sigler, at (619) 226-3881. You can also make an online donation by clicking here: Donate Now.

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