#### MARINE SKILL REPORT SUBMITTED TO THE UNIVERISTY OF HAWAII MARINE OPTION PROGRAM

### SALINITY, ITS RELATION TO BAITFISH

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#### INTRODUCTION

Topminnows have been used in Hawaii as a live commercial baitfish since the 1980's. Many species of <u>Poecilia</u> were captured by seine net in Honolulu and Pearl Harbor. They were then used by pole and line tuna fishermen. These fishermen captured topminnows out of necessity, because their traditional baitfish, the nehu (Stolephorus purpureus) was unavailable (Baldwin, 1980).

In Hawaii the nehu population is erratic and limited in quality (Brock and Uchida, 1968). Because of this, a cultured baitfish is essential if the tuna industry is to expand. On-going efforts are being made to raise topminnows as an alternate baitfish for the tuna fishermen.

Although fishermen prefer nehu, the topminnow has been used at FAD's (Fish aggregation devices), with success (DED, 1981), and appears the most suitable candidate for culture. In particular, <u>Poecilia mexicana</u> exhibits the most favorable characteristics of any topminnow tested thus far (Anonymous, 1981).

As a technician at the Maui County Baitfish Facility, I decided for m y Marine Option Program project skill, to study salinity and its effect on P. mexicana.

Although published information has dealt with salinity and its effect on <u>P</u>. <u>vittata</u> (Baldwin, 1977), an optimum salinity level for P. mexicana has never been explored.

Though Baldwin stated that the optimum salinity range is between 3.5  $^{\rm O}/\rm oo$  (parts per thousand) and 17  $^{\rm O}/\rm oo$  for <u>P. vittata</u>,

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no research has determined the optimum salinity range for <u>P. mexi-</u> cana.

<u>P. mexicana</u> has been raised in Western Samoa in salinities ranging from 0  $^{\circ}/_{\circ\circ}$  to 25  $^{\circ}/_{\circ\circ}$  (Popper, 1979). However, these ponds were filled by tidal fluctuations and salinity was not controlled.

At the Maui County facility, the salinity has ranged from 17  $^{\rm O}$ /oo to 20  $^{\rm O}$ /oo before the 1980 storm (Agres, 1983). Fry production for <u>P. mexicana</u> was considerably greater during the high salinity period (Anonymous, 1980). But due to numerous other variables, salinity could not be considered the limiting factor.

There are two main objectives to accomplish through this project. First, determination of a particular salinity level which enhances growth and survival rates for <u>P. mexicana</u>. Second, to test a higher salinity level then recommended, of 25  $^{\circ}/_{\circ \circ}$ . This is important to observe if <u>P. mexicana</u> exhibits similar rates of growth and survival at a wide range of salinities.

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#### MATERIALS AND METHODS

This study began with 3 circular pools. Each pool holds 2,000 gallons, is 12 feet in diameter, and 3 feet deep. One March AC-3C-MD 1/14 horsepower water pump draws 10 gpm (gallons per minute) out of each pool and into its own filter system. In each filter system the water flows through a Rainbow Lifeguard AF-94 mechanical filter, then into a Rainbow AF-93 chemical filter, then into a biofilter, and finally gravity flows back into the pool.

The mechanical filter has changeable filter paper to extract particulate material from the water. The chemical filter absorbs dissolved organic matter by passing water through crushed charcoal. The biofilter oxides ammonia waste from fish to less harmful nitrates (Spoote, 1970).

The biofilter consists of a 30 gallon plastic garbage can filled with course sand. A 3/4 inch PVC pipe with 1/16 inch holes drilled the length, is inserted into the bottom of the biofilter, then fastened across the diameter of the pool. The water flows from the biofilter, into the pipe, and streams across the pool.

Using a transport tank, I trucked ocean water from Maalaea harbor back to the project site. This was necessary due to the lack of a salt water well. I set the salinity in Pool 1 to  $4 \circ/00$ . This pool acted as the control and is similar to existing baitfish ponds. Pool 2 was set at 17  $^{\circ}/00$  and Pool 3 at 25  $^{\circ}/00$ .

I collected new born fry from the brood ponds. I passed them through a 3/32 inch eye net thus eliminating larger, older

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fry. I chose a stocking density of 10 fry per gallon. Baldwin suggests a stocking rate of 15 fish or less per gallon, so I used an intermediary rate.

The fry were fed tuna meal 3 times daily (morning, noon, late afternoon) at 5 <sup>O</sup>/oo of their total net body weight (Baldwin, 1977). Weight and length measurements were made bi-weekly and feed adjustments made according to growth.

Salinity was tested 3 times per week with a refractometer. Oxygen and temperature readings were taken 3 times weekly on a YSI oxygen meter. Readings were recorded for the surface and 2 foot water levels. Water quality testing was done bi-weekly consisting of ammonia, nitrite, nitrate, phosphate, and pH. These tests were performed on a Hach DR-EL4 test kit. Water was added to each pool to account for evaporation.

In the original proposal I stated the 3 pools would have plastic domes to increase temperature for optimum growth. The domes were in place 1 month before the experiment began. The plastic began to deteriorate after 3 weeks and tears appeared. Efforts to patch the tears proved futile as the tears would simply expand past the patch. I was forced to scrape the domes because of the expense of replacement plastic.

Another variation arose which was more satisfactory than the original proposal. I had intended to use lobster salt to increase salinties to desired levels. Fortunately, a transport tank became available and I was able to use ocean water. This allowed for a closer simulation to a brackish water well.

One final variation occurred in the water quality para-

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meters. Due to the salinity in the pools, the conductivity readings went off scale on the Hach kit. Thus no conductivity readings are available.

#### RESULTS

The following data was recorded over an 11 week period. The project was shortened from 16 to 11 weeks due to extensive fish mortality in Pool 1 and 2.

In week 1 the water in Pool 2 was milky white. For 2 weeks when cleaning the mechanical filter, I found a thick whitish gel-like susbstance collecting on the filter paper. By the end of week 2, the water had cleared up and the gel disappeared.

On August 13, I first noticed dead fry on the surface of Pool 2. On August 17 the water started turning from clear to dark and 2 days later, was black. I observed gas bubbles coming up from the bottom. On August 20, 4 weeks after stocking Pool 2, I could not detect any live fish. At this point, I discontinued monitoring Pool 2.

The nitrite, nitrate, and phophate levels in Pool 2 were negligible over the 1 month period. The ammonia, though reaching 0.38 mg/L, dropped back to 0.18 mg/L. Oxygen and temperature measurements were relatively constant except for a slight dip at the end of week 1. After 4 weeks, the average wet weight per fish in Pool 2 reached 0.027 grams. The mean average length reached 11.5mm.

On September 5, 7 weeks into the project, there was a slight drop in oxygen levels in Pool 1. This was followed 3 days later by a 5ppm upward swing. The next 2 oxygen measurements dropped

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and on September 14, were 4.3 ppm (surface) and 3.6 ppm (2 feet). Over the same period of time, the pool changed from green to brown and one day later, September 15, the water returned to a seemingly healthy green. From September 15 to the 18th, I was sick and unable to monitor the experiment. On September 18, I returned and found many fry dying in Pool 1.

The nitrite, nitrate, and phosphate levels were negligible throughout the cycle. However, ammonia levels shot up from 0.0 mg/L to 1.37 mg/L over a 2 week period and remained at that level. The pH was high from August 15 to the 30th when it reached 9.35. It then dropped to 8.12 on September 14.

On September 20, I recorded final weight and length measurements and discontinued monitoring Pool 1. The average wet weight per fish reached 0.081 grams. The mean average length was 17.88 mm.

For the next 2 weeks, I monitored Pool 3. The ammonia levels continued to climb throughout the experiment and the final measurement went off scale (>2.0 mg/L). Nitrite and phosphate levels were negligible, while pH remained consistent ranging from 8.0 to 8.5. Oxygen and temperature readings were consistent exhibiting no unusual fluctuations. At the end of ll weeks the average wet weight per fish in Pool 3 was 0.087 grams. The mean average length was 18.7 mm.

Due to the loss of the control (Pool 1), I discontinued the project at the end of week 11.

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#### DISCUSSION

When I first stocked the pools, the fry were released from the brood ponds (4  $^{\circ}/_{00}$ ) straight into Pool 2 (17  $^{\circ}/_{00}$ ) and Pool 3 (25  $^{\circ}/_{00}$ ). There was an initial mortality of 200 fry in Pool 2 and 500 fry in Pool 3. This might be attributed to transfer stress and the sudden change of salinity. After 2 days the population stabilized and further mortality was not observed until week 3.

The whitish gel began collecting on the filter paper of Pool 2 soon after the water turned milky. Efforts to identify the substance turned up nothing. I can only speculate that it was some type of algae but its consistency even leaves that possibility in doubt.

Upon cleaning out Pool 2 after discontinuation of its role in the experiment, I discovered a thick layer of detritus material covering the bottom. A rotten egg smell accompanied this material. The rotten egg smell is an indication of hydrogen sulfide. Rereading my observations made me suspect the gas bubbles that formed when the pool turned black might have been hydrogen sulfide gas (H2S). Un-ionized  $H_2S$  is extremely toxic to fish (Smith, 1976), and if the levels were high enough , it might have killed the fry. If the H2S levels were not high enough to kill the fry outright, the stress to the fry could have been enough to weaken the fry and allow disease to wipe the fry out (S.K. Johnson, 1976). I did not detect signs of disease, but my examination was limited to external observation.

In week 8 when the color in Pool 1 changed from green to brown, I suspected a phytoplankton crash. This crash could be related to lower oxygen levels (Boyd, 1975). The brown turbid

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color might be dead phytoplankton floating in the water column. The lack of healthy phytoplankton reduces photosynthesis and oxygen production.

In the same period of time that Pool 1 was experiencing strange color changes the amount of un-ionized ammonia (NH<sub>3</sub>) had reached 1.37 mg/L. NH3 levels of 1 mg/L and higher can be toxic to most aquatic organisms (Wheaton 1977). Low oxygen levels coupled with high NH3 levels can have adverse effects on fish populations (S.K. Johnson, 1976). Because many common fish diseases are a direct result of stress caused by depletion of dissolved oxygen (Aeration News, 1978), the topminnow fry may have become stressed to the point of susceptibility to disease.

Upon examination, the fry exhibited frayed fins and scale loss. This in itself is not conclusive evidence that fry in Pool 2 died from disease, but reviewing the events leading up to the mortality, I have to suspect disease caused by low oxygen levels.

When I first noticed lower oxygen levels, I should have been alerted to the possibility of oxygen depletion. At that point I should have switched on emergency aeration. I did not recognize the critical nature of the situation.

Due to mortality, I was unable to raise the minnows out for the recommended grow-out period of 4 months or 1 gram per fish baitfish size. Calculating the growth rates per day (0.0013 grams) per fish for Pool 1 over the shortened project, then projecting that rate over 120 days produces a 0.15 gram fish. This is only 15 percent of the suggested baitfish size. Either growth rates

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would greatly increase over months 3 and 4 or growth inhibitors were present in all three pools.

I was unable to find any information that dealt specifically with growth inhibitors. But on looking through Baldwin's final report (September, 1981), I came across a graph of stocking rates and subsequent growth rates. I compared my growth results (10 fish per gallon) with stocking rates of 5 and 7 fish per gallon and their respective growth rates (graph 10). From the graph it appears the lower the stocking density the greater the growth rate. If density is a limiting growth factor for topminnows, which it appears to be, then Baldwin's recommendation of 15 fish per gallon is too high.

Trying to draw conslusions from the limited data is difficult. From graph 9, it appears that <u>P. mexicana</u> grows faster in lower salinities. But since the rates were not markedly higher, <u>P. mexicana</u> could still be considered a candidate for marine polyculture. One advantage to the higher salinity is the ease of transition for the fish from the grow-out facility to an ocean water bait well.

If I did the same project over, I would use at least 6 pools. This would give me 2 replicates per salinity. If an emergency arose, I would want the ability to exchange water and try to flush the contaminant out of the system. This is also a closer approximation to pond situations. I was unable to exchange water because the closest salt water source was 3 miles away and the transport tank was no longer available.

I think it would be important to find out an optimum

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stocking density in future research. With that knowledge in hand, a duplication of this project with the preceding changes implemented, would be of value.

#### ACKNOWLEDGEMENTS

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Finally, I would like to thank Fred Matsumoto, Coordinator of the Maui County Economic Development Office for allowing me the use of the baitfish facilities.

#### EVALUATION

I chose to study salinity and its effects on topminnows for two reasons. First, because I was interested in aquaculture and thought what better way to learn more about aquaculture then to do a project on it. Second, I was employed as a baitfish technician at the Maui County baitfish facility.

I felt that for the use of the facility I should try to increase the information available on <u>P. mexicana</u>. Also, because the County was readying to begin a large scale baitfish facility. I thought the effects of salinity on topminnows might be significant as to the need for a salt water or fresh water well.

I discovered part of the step by step methodologies a scientist must involve himself with to assure valid results. I know now the importance of replicate information and how it can salvage a research project. I learned to deal with numerous small but important problems and details that crept up along the way. Difficulties in construction to data gathering techniques kept the project both interesting and frustrating all along the way.

I learned to use a Hach DR-EL-4 water quality kit. I developed crude methods of weighing and measuring fry the length of your thumb nail. And I watched pools die and wondered why.

Although the project failed in many ways, it was a very positive experience for me. I didn't learn a great deal about salinity and its effects on topminnows, but I did learn much about small tank culture.

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