Determining Dissolved Oxygen Tolerance of *Valamugil engeli* (Marquesan mullet) in Hawaiian Estuaries

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April 24th, 2014

Abstract

Juvenile fishes use estuaries for refuge, feeding, spawning, and adequate cover during critical growing stages. Shrinkage of an already small and physically constrained habitat may have significantly negative effects on water chemistry, and therefore affect the fish that locals have always relied on for subsistence. Mullet are popular recreational fish found year-round in estuaries and can tolerate large ranges of salinity. If water lacks the necessary amount of dissolved oxygen (DO) for a fish to survive, fish health and their populations will be affected. The goal of this experiment was to determine methodology for investigating dissolved oxygen tolerance, focusing on *Valamugil engeli* (Marquesan mullet). Juvenile mullet were collected from the Wailoa River and Hilo Bay estuary and tested for sub-lethal DO tolerance thresholds in a sealed jar. My results showed that Marquesan mullet have a significantly greater DO tolerance than expected. This information will assist in limiting mortality in Hawaiian mullet fisheries and provide utility towards developing simplistic experimental designs to test sub-lethal DO tolerance thresholds and metabolic rates of other estuarine fishes.

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I. Introduction

What is an Estuary?

An estuary is not easily defined, as many scientists have their own definition, each varying slightly from the next. Pritchard (1967) defines estuaries as ‘‘a semi-enclosed body of water which has a free connection with the open sea and within which sea water is measurably diluted with fresh water derived from land drainage.’’ This is one of the most widely accepted definitions. Another description that Odum (1995) uses is a, ‘‘river mouth where tidal action brings about a mixing of salt and fresh water.”

Estuaries have two main sources of water: groundwater and/or surface water. Estuaries are important for species diversity since they are very unique habitats of salt water and freshwater (Able 2005). These mixing zones allow amphidromous fishes to migrate between marine and freshwater habitats, such as mullets. Studies have shown that many juvenile fishes use estuaries for refuge, feeding, and adequate cover during critical growing stages, and some species then migrate to sea to spawn (Able et al. 1998; Hedgpeth 1982). As estuaries are impacted by coastal development, the high species diversity within these systems may also be threatened.

Estuaries in the Tropics

Many estuaries are located in the tropics (Blaber 2002). Tropical estuaries harbor a wide range of taxa that have close interactions within a single community. Furthermore, subtropical and tropical estuaries are known for high fish speciation compared to temperate regions (Blaber 2002). However, these estuaries tend to be characterized by challenging working conditions, resulting in little available research information for such areas, as study sites with more desirable working conditions are sought for (Blaber 2002).

Concern

One major concern is that Hawaiian estuaries are an exploited ecosystem (Blaber 2002). Some of the damage has been caused by increased coastal development, which adds to runoff, human activity pollutants, and decreased water quality. Rising sea level paired with decreases in freshwater discharge will limit estuary sizes. This shrinkage of what is already a relatively small and physically constrained habitat may have significant effects on water chemistry, and therefore the biota and ecological function of these systems. Many fish that locals have relied on for traditional fishing and subsistence, such as the mullet, could be affected if these changes to the estuaries become overbearing.

Mullet (Family Mugilidae)

Mugilids are a recreationally important fish throughout the world, and traditionally important in Hawai'i. Fisheries and aquaculture farms focus on rearing of these fish and enhancing wild stocks to ensure that numbers stay high enough to sustain the fishery (SHDLNR 2005). Mugilids are oblong silvery fish that travel in schools and spawn offshore. Juvenile mugilids feed on zooplankton and phytoplankton, whereas adults feed primarily on diatoms, detritus, and algae (Blaber 2002). In Hawaiian history, one species in particular, *Mugil cephalus*, known as *'ama'ama* in Hawaii, was respected by Hawaiian royalty and valued as a commercial product as far back as the 1900s. '*Ama'ama* are still highly valued as a food and game fish in Hawaii with a sizeable recreational fishery that exists across the islands.

Mullet represent a significant food source to upper level piscivores (Wenner et al. 1990). Mullet were chosen for the study species because they are widely distributed between latitudes 51^{o} N and 42^{o} S. Mugilids can tolerate large ranges of salinity and can be found year round (Harrison 2002).

The Marquesan mullet (*Valamugil engeli*) grows to about 18 cm total length in first year, after which they reach sexual maturity. Marquesean mullet typically grow no larger than 30 cm fork length. Distinct Marquesan mullet features include larger scales than the striped mullet, and a mouth that slants upward anterior to the eye (McDonough et al. 2003). The Marqusan mullet are considered alien because they were accidentally introduced to Hawai'i in 1955. The species quicky became established in estuaries and coastal regions throughout the island (Nishimoto et al. 2007).

Dissolved Oxygen

All fish use dissolved oxygen (DO) for respiration and are commonly very sensitive to its concentrations—too high and fish can developed air embolisms, which lead to mortality (Shehadeh et al. 1973); too low and fish will suffocate. If water lacks the necessary amount of DO for a fish to survive, it can cause fish to move into a new habitat or affect fish health and their populations. One way that low DO can cause negative effects was made evident by a study that showed fathead minnows that were tested in low DO concentrations over 11 months, which displayed a decreased number of eggs produced and frequency of spawning (Brungs 1971). It is speculated that DO is the cause of the differences in survival rates in captivity as researchers from the Oceanic Institute used low DO levels to sort juvenile striped mullets and the Marquesan mullets.

The purpose of this experiment is to better inform management decisions on the recreational shore fishery in Hawai'i, particularly the *'ama'ama* fishery, by understanding the physiology of its non-native competitor, Marquesan mullet. The goal of this experiment is to determine the DO tolerance of Marquesan mullet in Hawaiian estuaries. This experiment also provides insight into basic life history and biological information on this species, which will give better direction in estuarine riparian habitat restoration efforts in the future.

II. Objectives

My objectives are to investigate the DO tolerance level of the Marquesan mullet.

1) Designed experiment set up at station

2) Specimen collection

3) DO tolerance trials

4) Data analysis

III. Methods and Materials

<u>1) Designed experiment and set up at station</u>

Study Site

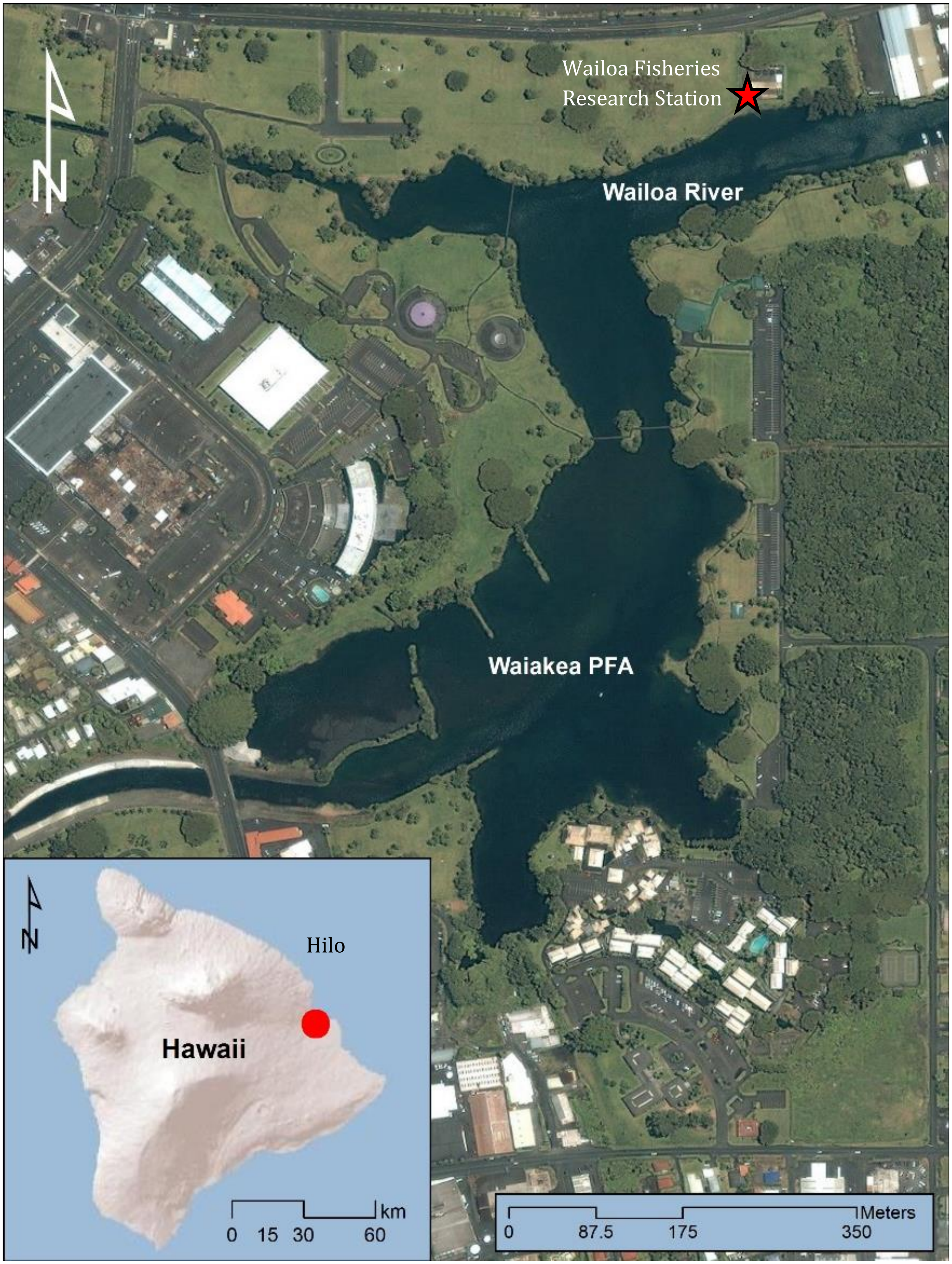

Figure 1- Map of Wailoa River collection site on the Island of Hawaii, USA.

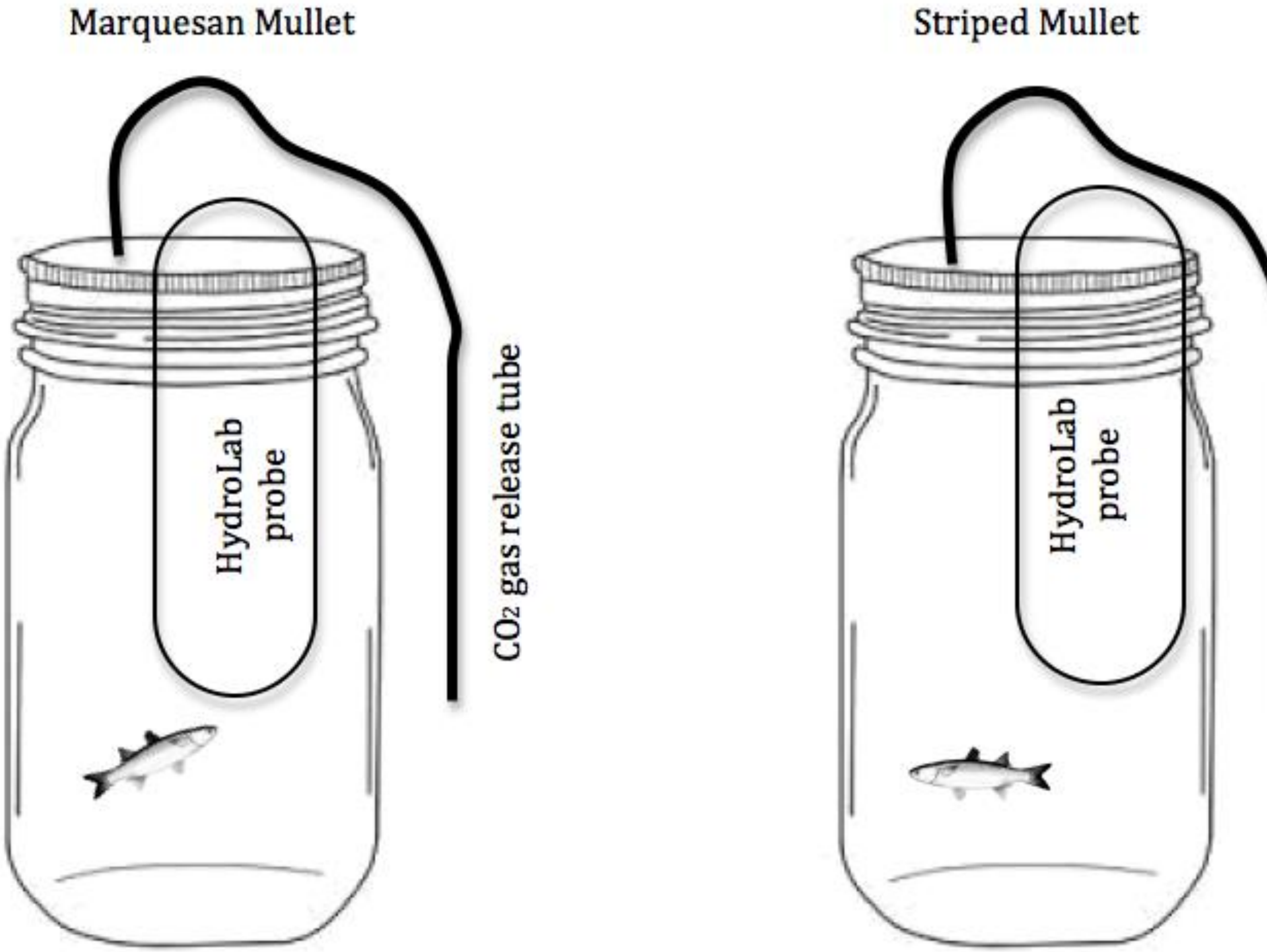

Figure 2- Dissolved oxygen testing design using 2120mL plastic jars, full with water and sealed, with a Hydrolab Quanta to measure DO concentration (mg/L) and DO % saturation, and CO_2 bleed tube.

Figure 3- Dissolved oxygen testing quarters with 5 fish and floating mesh, designed for this experiment

Figure 4- 378 liter water reservoir set up that saturated water to 100% and was used for recovery

Figure 5- 16 m^3 fiberglass housing tank

Figure 6- Floating net that was place in the housing tank to contain the juvenile fish

2) Specimen collection

This study was conducted in a tropical estuary, and the Wailoa Fisheries Research Station in Hilo on the Island of Hawaii. I conducted this study with the assistance and resources of the Division of Aquatic Resources (DAR). The role of DAR is to sustainably manage Hawaii's unique aquatic ecosystems and resources for present and future generations (State of Hawaii 2005). I used *Valamugil engeli* (Marquesan mullet) ranging from 47-101 mm fork length for this study. Specimens were collected from the Wailoa River and Hilo Bay estuary (Figure 1). Fish were collected with 0.6 cm mesh cast nets from shore and transferred back to the lab in covered buckets with aerator pumps on high to minimize stress and ensure survival.

The fish were housed in one 16 m^3 tank to ensure that all conditions were equal (Figure 5). Water in the housing tank was kept consistent with the conditions in Wailoa River in terms of water temperature, DO, and salinity levels. In order to do this, water was directly pumped in from Wailoa River using a Hayward Superpump II, which was run through a sand filter. The juveniles collected for the study were placed in a floating net to limit complications of collection for testing (Figure 6). Juvenile Marquesan mullet were fed fresh collected brown algae on Friday evenings, so the food would be out of the system by trial time on Monday. A reservoir of filtered Wailoa River water was kept in a separate 378 liter tank and constantly aerated to maintain 100% DO saturation (Figure 4). This water was used for recovery of the fishes after each trail.

3) DO Tolerance Trials

The dissolved oxygen tolerance was tested for juvenile Marquesan mullet by conducting fish trials in sealed 2120mL plastic jars that hold 2000mL (2 liters) with the displacement of the Hydrolab (Figure 3). Prior to the trials, 25 liters of filtered Wailoa River water was brought to 100% DO saturation using air stones (Figure 4). Fish were not fed for 24 to 48 hours prior to the experiment in order to clear their digestive tract, thus minimizing any confounding effects on metabolic rate from digestion. Five healthy Marquesan mullet were randomly selected from the holding tank and placed in the plastic jar design (Figure 3). Each fish was transferred as quickly as possible to minimize stress from handling.

A Hydrolab Quanta multimeter was placed through the top of the jar, and sealed with an o-ring. This Hydrolab displayed constant readings of DO percent saturation, DO (mg/L), and temperature (ºC). These readings were manually recorded in 2-minute intervals during each trial. Simultaneously, a control was run for that trial with a second HydroLab in an identical setup without fish. When the first juvenile fish showed loss of equilibrium (when the fish goes belly up, but gills still moving), all five fish were immediately removed from the jar and placed in a recovery bucket with 100% DO saturated Wailoa River water until full recovery. The Hydrolab Quanta multimeter was then stopped after final DO level and time it took to reach that point were recorded. After recovering, each tested fish was individually weighed (g) with a 400g balance and measured to fork length (mm) with a fish measuring board. Then, the fish were placed back into the recovery bucket with high DO. Once the fish fully recovered and were weighted and measured, they were released back into the Wailoa River estuary.

4) Data analysis

Graphs were made to view oxygen consumption rates and how long it took fish to reach disequilibration. The graphs show common anoxic level for marine fishes as well.

IV. Results

Experiment results unexpectedly showed that *Valamugil engeli* (Marquesan mullet) have a high DO tolerance range of 0.55-0.99 mg/L or 7.0-12.4%. All fish reached disequilibriation at

DO < 1mg/L.

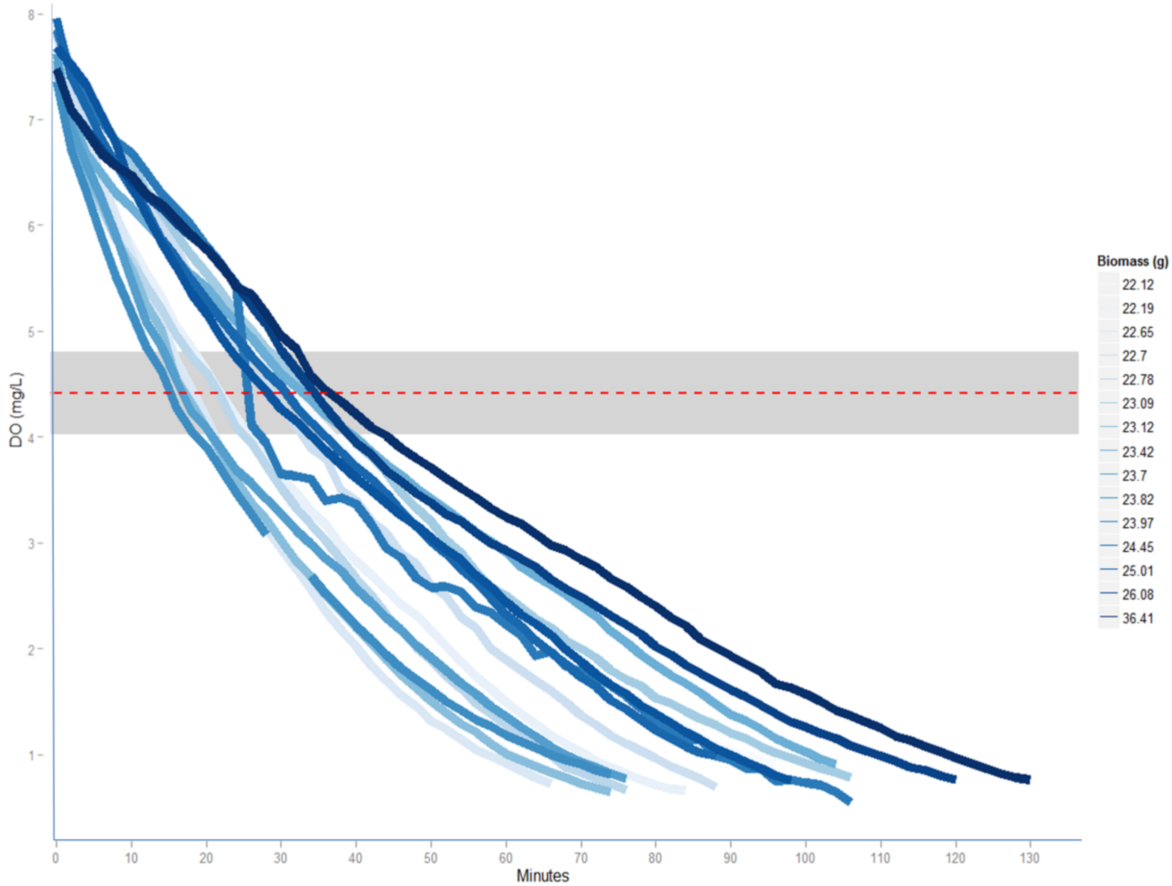

Figure 7- DO saturation changes over time until fish disequilibration for 15 trials, with color gradient representing masses. Red dashed line is the median sub-lethal hypoxic threshold for marine fishes (4.41 mg/L), the gray area is the standard error (± 0.39). These numbers are based on Vaquer-Sunyer and Duarte 2008.

V. Discussion

My research acts as a pilot study for DAR and other government fish agencies. Results show that in order to keep Marquesan mullet from loss of equilibrium when collecting from the field, or housing, DO mg/L must be kept above 0.99 mg/L or DO percent saturation above 12.4% (Figure 7). This study worked and was successful in providing the DAR with preliminary DO tolerance information for the Marquesan mullet.

We expected fishes to reach disequilibration around the hypoxic threshold for marine fishes. Levels reached in this study are far below the median sub-lethal hypoxic threshold for marine fishes (4.41 mg/L). Also, by looking at the masses, it is evident that the large masses of fish used up the oxygen faster than the fish of smaller masses (Figure 7). Speculation can also be made that temperature affects the rate of metabolism (Figure 7).

Conclusion

For future studies on DO tolerance experiments, I suggest that a smaller jar is used with only one fish, if possible. Size of probe to measure parameters must be taken into consideration. If they are schooling fishes being tested, a visual barrier must be used so they do not use cues from each other. The extended amount of time needed for water to get to 100% DO saturation needs to be considered. With 4 bubble blowers on high, it took overnight to saturate 378 liters of water. Care needs to be taken when housing the fish. Skin diseases or lesions need to be monitored for and fish may need to be treated with Methyl Blue. Any fish that do not appear normal should not be tested, but removed immediately and released into their natural environment.

This information better informs management decisions on the recreational shore fishery in Hawaii, more specifically the mullet fisheries. This experiment also provides insight into basic life history and biological information on the Marquesan mullet species. This gives better direction in estuarine riparian habitat restoration efforts in the future. Alien Marquesan mullet DO tolerance can be compared to the DO tolerance of the native Striped mullet in the future to see if one species has a competitive survival advantage amid the increasing impacts on and changes to the Hawaiian estuarine habitats. Findings can be used along with the project design to acquire DO tolerance level of many juvenile species to test varying parameters.

VI. References

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Investigations on the life history and population dynamics of marine recreational fishes in South Carolina, part 1. South Carolina Marine Resources Research Institute, Completion reports, Project F-37, Charleston and Project F-31, Brunswick.

VII. CV

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EDUCATION

Expected Graduation Date May 2014
Bachelor of Science in Biology with an emphasis in Ecology and Evolutionary Biology
Minnesota State University Moorhead (MSUM), Moorhead, MN
Spring 2014: 1 semester National Student Exchange at University of Hawaii at Hilo (UHH), Hilo, HI
GPA 3.81, Dean's List every semester

Relevant coursework: Organismal Biology, Principles of Ecology, Evolutionary Biology, Molecular Biology, Invertebrate Zoology, Vertebrate Zoology, Animal Behavior, Aquatic Biology, Quantitative Biology, Research Design, Tropical Field Biology (Costa Rica), Marine Coral Reefs (UHH), Wildlife Ecology, Physics, Marine Options Program Proposal (UHH)

RESEARCH EXPERIENCE

Spring 2014: Division of Aquatic Resources (DAR) 1160 Kamehameha Avenue, Hilo, HI 96720
Determining Dissolved Oxygen Tolerance of the Valamugil engeli *(Marquesan mullet) in Hawaiian Estuaries*
Mentor: Troy S. Sakihara

- Collected mullet from estuary via cast netting
- Set up and designed tank arrangement for testing of salinity and dissolved oxygen

Summer 2013: Auburn University, AL
Warm Water Ecology Research Experience for Undergraduates (REU), NSF funded
Hormonal and behavioral responses of Cyprinella venusta *(Blacktail shiner) to acoustic playbacks*
Mentor: Dr. Carol Johnston

- Collected Blacktail shiner in the tributaries of the Chattahoochee River
- Studied acoustic knocks and growls for fish and their meaning
- Practiced enzyme immunoassay procedure of free hormone extraction through water
- Performed outreach that educated daycare students with relevant ecological information

Spring 2011: MSUM
A method to train groups of predator-naïve fish to recognize and respond to predators when released into the natural environment
Mentor: Dr. Brian Wisenden

- Trained fathead minnows to associate alarm cue with predator scent and therefore associate danger
- Used to lower the mortality rate of fishes raised in hatcheries and released into the wild

Fall 2011: MSUM
Does Bt Corn Pollen Density Impact Monarch Butterfly (Danaus plexippus) Populations?
Mentor: Dr. Dan McEwen

- Studied the distribution of the pesticide Bt Corn pollen and effect on butterfly mortality

Fall 2011: MSUM
Genetic Mutations of Drosophila melanogaster
Mentor: Shireen Alemadi

- Studied the genetic lineage of mutations of fruit flies by breeding multiple generations

Fall 2011: MSUM
Invertebrates of the Prairie
Mentor: Becky Andres

- Collected and identified invertebrates for a pinned collection

PUBLICATIONS

Olson JA, **Olson JM**, Walsh RE, Wisenden BD. 2012. A method to train groups of predator-naïve fish to recognize and respond to predators when released into the natural environment. North American Journal of Fisheries Management 32: 77-81

PRESENTATIONS

Olson JM and Sakihara TS. 2014. Determining Dissolved Oxygen Tolerance of the *Valamugil engeli* (Marquesan mullet) in Hawaiian Estuaries. Poster presentation at the Marine Options Program (MOP) Annual Symposium in Oahu, Hawaii. Awarded best poster.

Olson JM. 2013. Hormonal and behavioral responses of *Cyprinella venusta* (blacktail shiner) to acoustic playbacks. Poster presentation at the REU capstone exhibit, in Auburn, Alabama.

Olson JA, **Olson JM**, Walsh RE, Wisenden BD. 2012. A Method to Train Groups of Predator-Naïve Fish to Recognize and Respond to Predators When Released into the Natural Environment. Poster at the National Animal Behavior Conference, Indiana University of Bloomington, July 2011

Olson JA, **Olson JM**, Walsh RE, Wisenden BD. 2012. A method to train groups of predator-naïve fish to recognize and respond to predators when released into the natural environment. Poster at the National Animal Behavior Conference, Indiana University of Bloomington, July 2011

Olson JM, Sluka KL, Kosak KJ. 2011. Does Bt Corn Pollen Density Impact Monarch Butterfly (*Danaus plexippus*) Populations? Poster presentation at the MSUM Student Academic Conference, April 2011.

Olson, JM, Sluka KL, Kosak KJ. 2011. Does Bt Corn Pollen Density Impact Monarch Butterfly (*Danausplexippus*) Populations? Poster presentation at the Student Academic Conference, Minnesota State University Mankato, April 2011.

SKILLS

Familiar Field Techniques

- Trap netting, backpack electro-shocking, cast netting, seining, mist netting, turbidity sampling, radio telemetry, limited knowledge in Spanish and sign language, proficient at driving manual

Data Analysis

- Familiar with analysis in R, regression and t-test in Excel, familiar with LoggerPro

Photographic Skills

- Underwater photography using GoPro camera, DSLR Nikon D200 portrait and nature photographer, familiar with Photoshop

Certifications

- Trained in CPR and AED, SCUBA, Safe Zone Training

WORK EXPERIENCE

Fall 2013

Teacher's Aid at Our Redeemer Christian Children's Center (ORCCC), Moorhead, MN

- Provided care for toddlers and infants
- Organized weekly crafts and engaging activities for toddlers to learn from

Summer 2012-Fall 2013

Dragon Ambassador, Admissions Office, MSUM

- Public relations representative for MSUM at alumni banquets, graduation, and presidential socials
- Informed prospective students of opportunities while giving campus tours

Summer 2012

Cocktail and Food Waitress at the Buffalo Wallow in Sisseton, SD

- Responsible for bussing tables, taking and calculating bills, running the till, serving, prep cooking, and dish washing

Summer 2011

Prairie Sky Guest and Game Ranch –Girls' Horse Camp Counselor in northeastern South Dakota

- Provided teenage girls with guidance and information on western horseback riding
- Taught teenage girls to care for horse and tack by cleaning and grooming
- Organized nightly crafts or activities for camp attendees
- Responsible for supervising the camp attendees while staying in a cabin

April 2011-May 2013
Resident Assistant, Housing and Residential Life, MSUM

- Acted as a leader and mentor for students living on campus and coordinated programs
- Completed intense paperwork on time as required by campus policy
- Executed leadership skills through serving in multifaceted supervisory role
- Capable of making decision, customer service, and confronting people on the spot

Summer 2008-Summer 2012
American Red Cross Certified Lifeguard and Water Safety Instructor, SSPA Sisseton, SD

- Supervised and provided first aid to customers
- Developed and partook in engaging lesson plans for swimming lessons for Preschool to Level 6

Summer 2006 and Summer 2007
Car Detailer at Service Plus Auto Repair Shop in Watertown, SD

- Vacuumed car interiors
- Washed car interior and exterior windows
- Responsible for timely car delivery to customers

EXTRACURRICULAR ACTIVITIES AND HONORS
Fall 2013
Elected Ms. MSUM (Mock Pageant)

- Preformed talent, runway, and short answer portion of pageants

Fall 2013
Co-host of 4th annual Girls' Night, Breast Cancer Awareness Event, MSUM

- Held and organized meetings
- Delegated specialized committees
- Contacted breast cancer survivor to share a story of her struggle and importance of early detection
- Donated money raised to Bras on Broadway event held in Fargo, ND each year

Spring 2011-Fall 2013
Historian for the National Resident Hall Honorary (NRHH), MSUM

- Partook in monthly community service
- Took photos at community service and volunteering events
- Managed social media by providing updates of events and service projects

2010-2012
Wildlife Society, MSUM

- Helped organize volunteer projects on Zoo Enrichment
- Assisted in making t-shirts

Spring Break of 2012
Students Today. Leaders Forever (STLF), North Dakota State University,

- Attended "Pay it Forward" Trip over spring break to Texas, performed service projects
- Donated time and talent to multiple non-profit organizations on the way to Dallas, Texas

Fall 2010-Spring 2011
Resident Hall Association, Snarr Hall Representative**,** MSUM

- Represented students who live in the residence halls by being student voice of a hall
- Assisted in organizing, advertizing, and planning campus wide events

Selected as Senior Snow Queen, Sisseton, SD

- Participated in entry walk, formal, and interview section
- Represented my home town as Ms. Sisseton at the state pageant

SMALL BUSINESS EXPERIENCE
Olson Photography
http://olsonphotographywebstartscom.WebStarts.com/
https://www.facebook.com/jolsonphotography

- Experience in creation, scheduling, budgeting, and complete management of a small business

REFERENCES

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