# Laboratory Growth Responses of Juvenile Mugil sp. to Temperature and Salinity: Delineating Optimal Field Growth Conditions

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#### ABSTRACT

Mullet (Mugilidae) are distributed worldwide in temperate and tropical marine environments, are eurytolerant of variable conditions, are commercially exploited, and have been used as a model for marine stock enhancement. Given this eurytolerance and the apparent decline of mullet in the northern Gulf of Mexico, we were interested in quantifying the influence of temperature and salinity on growth as it relates to the determination of optimal field growth conditions. We grew young juvenile mullet in a randomized and interspersed 3x4 factorial design (20, 25 and 30°C and 3, 10, 17 and  $24^{\circ}/_{00}$ ) with nine replicates each (five fish/replicate) over a 30 day period. Results from the laboratory experiments revealed significant temperature (p < 0.001) and salinity (p = 0.019) effects on growth, with no interaction term (p = 0.964). These data suggest optimal growth occurred at temperatures ≥ 25°C, and, within each temperature treatment, peak growth occurred at 170/co. To compare these results to growth in the field, modal shifts in length-distributions of recruiting cohorts of young juvenile mullet were considered in relation to continuous changes in ambient abiotic conditions monitored with Hydrolabs at two widely-separated locations (45 km apart) along the Mississippi coast. Modal standard length change of young juvenile mullet over a seven day period was 3.4 mm (0.486 mm/d) at the Marsh Point location and was 2.2 mm (0.314 mm/d) at the Henderson Point location over the same time period. This is a 35.4% difference in standard length over seven days, which when coupled with the salinity and temperature data noted above, parallel and generally support the differences observed from the laboratory growth experiments.

KEYWORDS: Abiotic, growth, Mugilidae

#### INTRODUCTION

Mullet (Mugilidae) are distributed worldwide in temperate and tropical marine environments, are eurytolerant of variable conditions, are commercially exploited, and have been used as a model for marine stock enhancement (Leber and Arce 1996). Both striped (Mugil cephalus) and white (M. curema) mullet spawn offshore in the northern Gulf of Mexico. Larval striped mullet are

abundant from October to March while larval white mullet are abundant from April to mid-September (Ditty and Shaw 1996). Young of both species recruit into estuarine habitats where they are exposed to variable abiotic factors that may influence their survival and growth (Marais 1978, Nordlie et al. 1982). For example, laboratory experiments with striped mullet 20-39 mm standard length (SL) indicated they were able to tolerate instantaneous transfer from brackish water (6-23°/ $_{\odot}$ ) at 22° C to salinities up to full strength seawater (Nordlie et al. 1982). However, calculated energetic cost of osmoregulation of striped mullet (O weight = 15-18g @ 25° C) was high when salinity was hyperosmotic with respect to the blood isosmoticity (~13-15°/ $_{\odot}$ ), Nordlie et al. 1982); the cost was negligible when the environmental osmolality was less than or equal to that of the blood (Nordlie and Leffler 1975). White mullet (23-97 mm total length (TL)) have a high survival rate at 8.5°/ $_{\odot}$ 0 in temperatures between 13-30° C and are most stressed at 1.7°/ $_{\odot}$ 0 or 34°/ $_{\odot}$ 0 than at intermediate salinities at this size (Fanta-Feofiloff et al. 1986).

Estuaries are characterized by pronounced spatial and temporal variation in physical-chemical conditions which can directly or indirectly influence survival and growth of a number of estuarine-dependent fishes (Malloy and Targett 1991, Neill et al. 1994, Lankford and Targett 1994) due to lethal or stressful conditions. These conditions can influence or constrain the relative value of estuarine nursery zones but usually are not considered in the delineation of Essential Fish Habitat (EFH) for juvenile fishes. The objectives of this study were

- i) to estimate laboratory growth of juvenile *Mugil* sp. exposed to twelve temperature/salinity combinations, and
- ii) to relate these laboratory growth data to field growth data from two estuarine locations along the Mississippi coast where fish experienced different environmental conditions.

These data are considered relative to delineating optimal field growth conditions which is vital to establishing EFH for mullet.

### MATERIALS AND METHODS

#### Field Collection Procedures

Fish for the laboratory experiments were collected with a bag seine constructed of 3.2 mm mesh between 16 and 19 May 1997 from Biloxi Bay, Mississippi (Figure 1). Fish were transported to the laboratory in coolers, transferred into holding aquaria (see below) and later sorted by size. Although there is limited overlap between spawning seasons of striped and white mullet, both species were present in our spring collections. Because these fish were small and could only be separated by counting the number of anal elements

(Ditty and Shaw 1996), which would require examination under a microscope, we opted to examine the growth of young mullet at the generic level. Field collections at Marsh Point (n = 5) in Jackson County and Henderson Point (n = 5) in Hancock County (Figure 1) were made between 12 May and 16 June 1997 in order to estimate modal shifts in length-distributions of recruiting cohorts of young juvenile mullet relative to continuous changes in ambient abiotic conditions. All field caught mullet were measured for SL with calipers to 0.1 mm. Ambient abiotic conditions were monitored with Hydrolab IV recorders at Henderson Point and Marsh Point, two locations situated 45 km apart along the Mississippi coast that are primarily influenced by different watershed systems.

## Laboratory Procedures

Fish were maintained at  $24 \pm 1^{\circ}$  C and  $13^{\circ}/_{00}$  under flow-through conditions (total volume exchange every 6 h) in two fiberglass aquaria (~216 and 330 L) until 3 June 1997 (15-18 days). Five fish (< 25 mm;0 wet weight (WW)= 2.89g) per replicate were then transferred to experimental aquaria (see below) for six additional days. These preliminary periods allowed for any mortality prior to experiment initiation. Fish from each replicate aquarium were removed on 9 June 1997, weighed (WW to 0.001g) in groups of five on an Ohaus Balance, and returned. During maintenance and experimental periods fish were fed thawed and rinsed brine shrimp ad libitum twice daily for 10 minutes and then all remaining food was removed. Fish were not fed on the days they were weighed. After 30 days, fish from each replicate were re-weighed and placed in 95% ethanol.

Water temperature in each replicate aquarium was maintained in three connected water baths arranged in a vertical stack for each experimental temperature (20, 25 and 30°C, n = 9 baths). Thermostatically-controlled 1-kw submersible heaters and a Frigid-Unit water cooler were used to maintain water temperature in the air-conditioned laboratory. Water temperature of each bath was recorded daily, and salinity was recorded about every other day from each replicate tank beginning 3 June and ending at the close of the experiment. Each replicate consisted of a 21 L glass aquarium supplied with saline water pumped from floor-vaults containing the appropriate salinity. Water was completely exchanged in each experimental aquarium every 24 hours and water level was maintained with an external standpipe that siphoned water from the bottom of each experimental aquarium.

We grew young juvenile Mugil sp. in a randomized and interspersed 3x4 factorial design (20, 25 and 30°C and 3, 10, 17 and  $240/_{\infty}$ ) with nine replicates each (five fish/replicate) over a 30 day period. Because we could not follow individual growth within each replicate, we calculated the mean relative increase in body weight (Ricker 1975) as  $G = ((\log_{10} \text{ FWWM-log}_{10} \text{ IWWM})/\log_{10} \text{ IWWM})$ 

IWWM) where FWWM = mean final WW and IWWM = mean initial WW. This growth estimate served as a response variable in a two-way Analysis of Variance (ANOVA). If a significant F-value (p < 0.05) was obtained, a Sidak pairwise comparison test was used to distinguish treatment means. All tests were conducted with SPSS (Windows Version 7.5) statistical software (SPSS, Inc., Chicago, Ill).

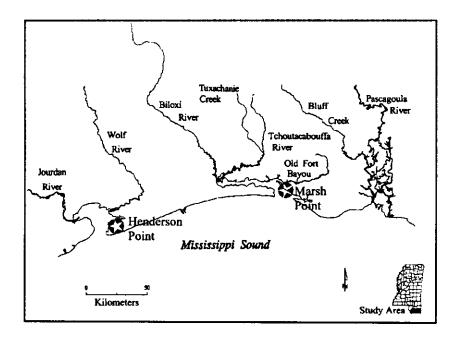
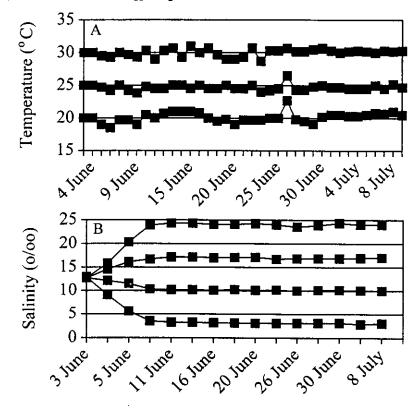


Figure 1. Map of the field sampling locations in coastal Mississippi.

#### RESULTS

# Laboratory conditions and growth

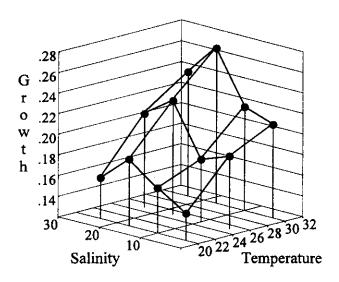
Over the course of the experimental period, temperature and salinity were consistent with desired conditions (Figure 2). Of the 510 fish used in these experiments, 64.1% were striped and 35.9% white mullet. Significant temperature (p < 0.001) and salinity (p = 0.019) effects on growth were found, with no interaction term (p = 0.964; Table 1). These data indicate optimal growth of juvenile *Mugil* sp. occurred at temperatures  $\pm 25^{\circ}$ C (Sidak,  $20 < 25 = 30^{\circ}$ C), while within each temperature treatment, peak growth occurred at  $17^{\circ}/_{\infty}$  (Sidak,  $3 = 10 < 17 > 24^{\circ}/_{\infty}$ ) (Figure 3).



**Figure 2.** Plot of mean temperature (A) and mean salinity (B) for the laboratory experiments. Temperature values are based on the daily values of the three water baths and salinities are based on the nine replicate values taken 15 times (~ every other day) during the experimental period. The experiment began on 9 June.

**Table 1.** ANOVA summary table comparing G ((log<sub>10</sub> FWWM-log<sub>10</sub> IWWM)/  $log_{10}$  IWWM) of *Mugil* sp. exposed to a 3 x 4 factorial experiment with temperature (20, 25, 30°C) and salinity (3, 10, 17, 24°/ $log_{10}$ ). FWWM = mean final wet weight, IWWM= mean initial wet weight.

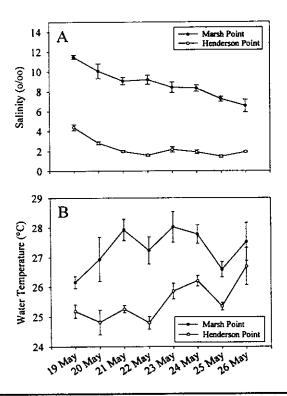
Source	df	Sum of Squares	Mean Squares	F	p
Intercept	1	4.390	4.390	1267.384	.000
Temperature	2	0.00989	0.00495	14.278	.000
Salinity	3	0.00362	0.00121	3.488	.019
Temperature *Salinity	6	0.00049	0.00008	0.236	.964
Error	96	0.33300	0.00035		
Total	108	4.863			



**Figure 3.** Plot of the mean relative growth (( $\log_{10}$  FWWM- $\log_{10}$  IWWM)/ $\log_{10}$  IWWM) of young juvenile *Mugil* sp. exposed to a 3x4 factorial experiment with temperature (20, 25, 30°C) and salinity (3, 10, 17, 24° $\log_{10}$ ) as the main factors.

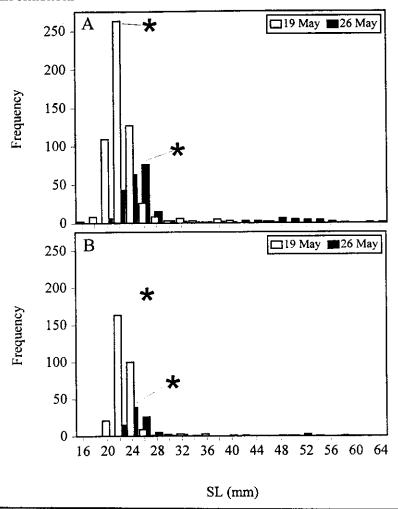
### Field conditions and growth

Daily mean salinity conditions at the Marsh Point location declined from a high of 11.47 to 6.540/00 between 19-26 May 1997 while at the Henderson Point location, mean salinity declined across a lower range from a high of 4.39 to 1.580/00 (Figure 4A) over the same time period. Daily mean water temperature ranged from 26.16 to 27.53°C at the Marsh Point location while at the Henderson Point location it ranged from 24.81 to 26.70°C (Figure 4B). Clearly, the salinity was higher and much closer to the optimum of 170/00 observed in laboratory experiments at the Marsh Point location compared to Henderson Point. Mean water temperature between locations differed by a maximum of 1.3°C, but relative fluctuations were similar at both locations (Figure 4B).



**Figure 4.** Plot of daily ( $\bar{x} \pm 2$  SE) salinity (A) and water temperature (B) at Marsh Point and Henderson Point between 19-26 May 1997. Plots are based on hourly values each day obtained from HydroLab IV units.

The 19th and 26th of May 1997 were the first two dates when young juvenile *Mugil* sp. were collected in sufficient numbers at both locations to estimate modal shifts in SL (mm) (Figure 5). At Marsh Point (Figure 5A), the modal SL changed from 21.8 to 25.2 mm over 7 d (3.4 mm; 0.486 mm/d) while at the Henderson



**Figure 5.** Length-frequency plot of young juvenile *Mugil* sp. on 19 and 26 May 1997 at the Marsh Point (A) and the Henderson Point locations (B). \* = modal SL. Sample sizes for Marsh Pt. were 568 ( $19^{th}$ ) and 245 ( $26^{th}$ ). At Henderson Pt. sample sizes were 303 ( $19^{th}$ ) and 98 ( $26^{th}$ ).

Point location (Figure 5B) the modal SL changed from 22.4 to 24.6 mm over the same time period (2.2 mm; 0.314 mm/day). This is a 35.4% difference between the two locations. These changes in SL over 7 day, coupled with the salinity and temperature data noted above, parallel and generally support the differences estimated from the laboratory growth experiments. Physical data suggest salinity differences between locations could have primarily driven growth differences in mullet, although differences in water temperature might have reinforced the growth difference.

### DISCUSSION

Defining physical-chemical conditions where optimal growth occurs in juvenile fishes is vital to understanding the relative value of nursery habitats and is an essential component in delineating EFH. Field growth at two locations along the Mississippi Gulf coast generally paralleled our laboratory delineation of optimal growth conditions in young juvenile *Mugil* sp. Growth in the laboratory was influenced significantly by temperature and salinity suggesting that growth of individuals recruiting into habitats with similar differences in these physical-chemical conditions should differ accordingly in growth, assuming other potentially confounding factors such as food availability at each location were equal.

It is unlikely that our laboratory and field growth responses were confounded by the mixed nature of the specimens used due to species- or size-specific differences in salinity and temperature tolerance. For example, the respiratory metabolism of striped and white mullet is similar at temperatures between 15-25°C, although respiratory metabolism increases in white compared to striped mullet at higher temperatures (Moore 1976). Our laboratory data indicate however that young juvenile Mugil sp. grew faster at temperatures ± 25°C and at salinities of 17% on Indeed, maximum growth in our laboratory experiments was very similar to the estimated 13-15% isosmotic point in striped mullet (Nordlie et al. 1982) where energetic costs are lowest. If temperature and salinity tolerance influenced striped and white mullet growth very differently, we would expect to see variable and confusing results. Instead we documented clear statistical differences among treatments. Additionally, since temperatures between the two field locations varied by a maximum of 1.3°C on average and were above 25° C during our collections, it is unlikely that this minor difference differentially influenced field growth rates of the mullet from the two locations.

Salinities were markedly different between the two locations and, assuming other potentially confounding factors such as food availability at each location were similar, have influenced growth of young juvenile mullet. Thus, the small size of the fish used in this study and the environmental conditions at the time

of collection indicate estuarine conditions are ideal for young juvenile mullet but that growth can vary over the scale we examined. These data are supported by studies which show an ontogenetic pattern to salinity tolerance in striped and white mullet. For example, optimal hatch rate conditions of striped mullet are  $36.3^{\circ}/_{00}$  at  $25.5^{\circ}$  C with the temperature tolerance limit for normal development being above  $30^{\circ}$  C; hatch rate varied with embryo developmental stage (Walsh et al. 1991). Nordlie et al. (1982) showed that the ability to tolerate lower salinity increased with body size and that young juvenile mullet (20 - 29 mm SL) could not tolerate freshwater until they were larger than 40 mm SL. A similar salinity tolerance pattern was determined for young juvenile white mullet as well (Fanta-Feofiloff et al. 1986). In addition, juvenile striped mullet (10g) exhibit a lower metabolic rate across temperatures ranging from  $13 - 33^{\circ}$ C in  $1^{\circ}/_{00}$  than in  $35^{\circ}/_{00}$  and a lower metabolic rate than 100g striped mullet under similar experimental conditions (Marais 1978).

Mullet are distributed in temperate and tropical marine environments worldwide and are commercially important throughout the Gulf of Mexico (Leard et al. 1985). In Hawaii (Leber and Arce 1996), striped mullet have been used as a model for marine stock enhancement. As part of the validation of the striped mullet stock enhancement program, Leber and Arce (1996) stocked cultured striped mullet (45-130 mm total length) into three locations over three years in Kaneohe Bay, Hawaii and followed them into the commercial fishery. Over the three years of this effort they showed that striped mullet grew differentially among years at a single location and among the three locations within years (Leber and Arce 1996). They also determined that fish < 60 mm TL had the greatest survival when released in spring (5-17 May) compared to summer (12-26 July) (Leber et al. 1997). Survival may have been influenced by differences in ambient temperature and salinity at each location during their study. Unfortunately, neither study provided temperature and salinity data by location or year. Given our results with young juvenile mullet, it might be expected that the physical-chemical conditions at any release location may have influenced growth and subsequent survival.

Juveniles of many species of fish require estuarine habitat that is characterized by large spatial and temporal fluctuations in physical and chemical conditions. Our study showed how these variable abiotic conditions can influence growth and potential survival of juvenile mullet. This kind of data can be useful in developing a more complete understanding of factors influencing recruitment variability and, from a practical point of view, can assist in improving survival of stocked marine fishes.

#### ACKNOWLEDGMENTS

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