# The Impact of Pre-Release Exposure on Survival Success in Hatchery-Reared Fish

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## ABSTRACT

The success of stock enhancement programs is contingent upon survival of hatchery-reared progeny. In this study, we investigate the effect of pre-release exposure on the survival of a marine finfish, red drum (*Sciaenops ocellatus*). Red drum larvae were collected from a Texas Parks and Wildlife Department (TPWD) hatchery and reared with and without the presence of natural vegetation (*Spartina alterniflora*) for either 10 or 20 days High-speed video was used to analyze a suite of prey-capture performance and anti-predator response variables at day 28 (~30 mm TL) and day 38 (~40 mm TL). Multivariate testing indicated that overall performance of juvenile red drum increased with age but rearing habitat did not enhance survival skills. Univariate contrasts showed that timing and distance of key response variables (e.g. maximum gape, time to maximum gape, gape cycle duration and time to maximum velocity) increased significantly with age. Although the multivariate models indicated that exposure to natural vegetation did not influence overall performance, univariate contrasts showed that time to reach maximum velocity was faster for individuals reared in vegetation; however, these fish also had a shorter reaction distance to predators and took longer to capture prey. Our results demonstrate that feeding performance and anti-predator response increases with age, while exposure to natural vegetation does not appear to afford any obvious benefits. Work addressing the effect of predator exposure on red drum performance is currently underway and will also be discussed in the context of supplemental stocking.

KEY WORDS: pre-release exposure, stock enhancement, red drum

# El Impacto de la Exposición Previa a la Partida en el Éxito de la Sobrevivencia en Peces de Cultivo

El éxito de los programas para el mejoramiento de las reservas de Corvinas [Red Drum] depende de la sobrevivencia de la descendencia hecha en criaderos de peces. En este estudio, investigamos el efecto de la exposición previo a la partida en el éxito de la sobrevivencia de un pez marino con aleta, Corvina [Red Drum] (*Sciaenops ocellatus*). Las larvas de Corvinas [Red Drum] fueron extraídas de un criadero del Texas Parks and Wildlife Department (TPWD) y luego criadas con y sin la presencia de vegetación natural (*Spartina alterniflora*) sea por 10 o 20 días. Video de alta velocidad fue utilizado para analizar un conjunto de variables de la eficacia de captura de presa y la respuesta contra depredadores en el día 28 (~30 mm TL) y en el día 38 (~40 mm TL). El examen estadístico de múltiples variables indicó que el rendimiento general de Corvinas [Red Drum] jóvenes incrementó con la edad, pero que el hábitat de crianza no tuvo ningún efecto en las habilidades de sobrevivencia. Contrastes univariables demostraron que el tiempo y distancia de variables de respuesta claves (apertura bucal máxima, tiempo a la apertura bucal máxima, duración del ciclo bucal y tiempo a la velocidad máxima.) aumentaron significativamente con la edad. Aunque los modelos multivariables indicaron que la exposición a la vegetación natural no influenció el rendimiento en general, contrastes univariables sí mostraron que el tiempo para alcanzar máxima velocidad era mas rápido para individuos criados en hábitat con vegetación natural; sin embargo, estos peces también tenían una distancia de reacción más corta a los depredadores y tardaron más tiempo en capturar la presa. Nuestros resultados demuestran que la eficacia de la alimentación y la respuesta contra depredadores aumenta con la edad, mientras que la exposición a la vegetación natural no parece producir ninguna ventaja aparente. Trabajo que trata con el efecto de la exposición a depredadores en el rendimiento de Corvinas [Red Drum] está actualmente en curso y será discutido en el contexto de mejoramiento de reservas suplementario.

PALABRAS CLAVES: exposición al prelanzamiento, mejoramiento de reservas, Corvina

## INTRODUCTION

Red drum historically supported a thriving commercial fishery in the Gulf of Mexico, and large-scale stock enhancement programs for red drum currently exist in several states in the U.S. (Florida, Texas, South Carolina, Georgia) (Woodward 2000; Smith et al. 2001). Although supplemental stocking of red drum has been linked to the recovery of natural populations (McEachron et al. 1995; McEachron et al. 1998; Jenkins et al. 2004), previous studies have shown that hatchery red drum experience lower growth and higher mortality than their wild counterparts (Rooker et al. 1998; Stunz et al. 2002a), and this may be linked to their ability to effectively utilize complex habitats (Stunz et al. 2001). Moreover, recent evidence suggests that the behavioral performance (e.g. survival skills) of hatchery-reared red drum is diminished relative to wild individuals (Smith and Fuiman 2004). As a result, hatchery red drum may lack critical survival skills, which may lower post-release survival and reduce the effectiveness of current supplemental stocking initiatives.

using a suite of variables previously linked to survival success. This study represents the first attempt to determine whether habitat exposure is linked to early life survival in hatchery-reared fish, and our findings will complement other efforts aimed at assessing the relative value of pre-release conditioning measures (e.g. exposure to predators) for red drum.

## METHODS

### Habitat exposure protocol

For our experimental trials we used 18-day old red drum larvae (10-12mm SL) collected from a single rearing pond from the Texas Parks and Wildlife Department SeaCenter hatchery in Lake Jackson, Texas. This size range was chosen since it parallels the size when wild individuals have fully recruited to estuarine nursery habitats (Rooker and Holt 1997). To assess the effect of habitat on performance, red drum from TPWD were transferred to our experimental rearing facility in Galveston, Texas and stocked into fiberglass tanks (1.5 m diameter, 0.75 m deep, 35 fish/tank) with and without vegetation (4 replicates per treatment = 8). Bottoms of each tank contained approximately 10-cm commercial washed sand and for our vegetated treatments we planted smooth cordgrass (*Spartina alterniflora*), a common nursery habitat for red drum (Baltz et al. 1993; Stunz et al. 2002b), into each tank at a density comparable to natural stem densities (~ 100 stems/m$^2$, Gleason et al. 1979). Red drum were fed a mixture of mysid shrimp (*Mysidopsis* spp.) and enriched 2-day *Artemia franciscana* once daily throughout the course of the trial. Water changes (20%) were performed several times weekly using filtered seawater pumped from the Gulf of Mexico and lighting was provided by fluorescent bulbs (10-12 $\mu$ E s$^{-1}$ m $^{-2}$) placed on a 12L:12D cycle to simulate natural light conditions. At the end of a 10-day rearing period, red drum were sampled randomly from each tank and placed into individual chambers (18 cm x 10 cm) for high-speed video analysis. Final mean standard lengths (SL) of individuals were 24.25 ± 0.104 mm and 23.04 ± 0.114 mm for vegetated and non-vegetated tanks, respectively, corresponding with historical TPWD release sizes (McEachron et al. 1998).

### Prey-capture performance

Six prey capture response variables were quantified: (1) attack distance (distance from the tip of the premaxilla to the closest point on the prey at the beginning of prey-capture), (2) mean attack velocity (average red drum velocity from time zero to when prey enters the mouth), (3) time to prey-capture (time to when prey enters the mouth), (4) maximum gape (distance from tip of the premaxilla to the tip of the dentary bone), (5) time to maximum gape (time when maximum gape is reached), and (6) gape cycle duration (time elapsed from initial mouth opening to mouth closing). Prey-capture performance was evaluated by recording a series of red drum feeding strikes on mysid shrimp (3-5 mm), a natural prey item of red drum at this stage (Soto et al. 1998).

### Anti-predator response

Immediately following prey-capture trials, anti-predator response of red drum was assessed using a visual stimulus consisting of a 4.5-cm diameter bulls-eye target on a swinging pendulum arm. This stimulus was modeled after a similar experiment by Batty (1989) and has been shown to effectively produce an escape response in red drum larvae and juveniles (Fuiman and Cowan 2003; Smith and Fuiman 2004). All anti-predator trials were conducted in a separate control box to minimize the effect of observer influence, and fish were allowed to acclimate for 20 minutes before the stimulus was introduced. The pendulum was released by the observer when the fish was near the front of the chamber and pointing towards the direction of the stimulus. Six variables were recorded for red drum, including: (1) reaction distance (distance between red drum and center of target at time zero), (2) distance traveled during the first 100ms of response, (3) maximum velocity reached during response, (4) time to maximum velocity during response, (5) mean velocity during response, and (6) maximum acceleration attained during response. Filming began simultaneously with the release of the stimulus and was blocked from making contact with the chamber. During a typical response, fish bent sharply to the right or left away from the approaching stimulus (C-start; Eaton et al. 1991) and swam rapidly towards the opposite end of the container. In many cases, we were unable to analyze an entire anti-predator response since fish either made contact with the sides of the container or swam outside the field of view during the course of an escape event. Therefore, we analyzed only the first 100ms of each response and used this data in our final analysis.

### High-speed videography

Prey-capture performance and anti-predator response of each red drum was filmed at 250 frames sec$^{-1}$ using a Redlake MotionScope PCI-1000s high-speed video camera. Prey-capture events were filmed laterally to the camera and anti-predator responses were filmed from above. A 1cm x 1cm grid placed behind the control box was used to provide scale for high-speed video analysis. Responses of three red drum were evaluated from each tank, and an average of three successful prey-captures and anti-predator responses for each fish were recorded. Redlake MotionScope and Peak Motus software were used to analyze prey-capture and anti-predator performance. Each variable was referenced to 'time zero', corresponding to the frame prior to mouth opening during feeding and the frame immediately preceding the first movement away from the stimulus during an anti-predator response. Velocity and acceleration data were calculated by tracking a digitized point on the center of the eye during prey-capture and the center of mass (~ 30% of body length from tip of snout, verified from preserved specimens) during anti-predator response. Displacement data for velocity and acceleration estimates were uploaded to QuickSAND software for MAC and smoothed using a generalized cross validation (GCV) quintic spline (Walker 1997). A second quintic spline with a previously determined mean squared error (MSE, 0.0001) produced similar results to the GCV quintic spline and is therefore not reported.

**Data analysis**

Multivariate analysis of variance (MANOVA, Pillai's Trace) was used to test the null hypothesis that survival skills did not significantly differ between red drum reared in vegetated versus non-vegetated tanks. Univariate contrasts were generated separately for prey-capture performance and anti-predator response variables. All tests were performed on tank means, which were calculated by averaging the response variable of three fish sampled from each tank. In addition, we used repeated-measures analysis of variance (ANOVA) to test for differences between individuals since fish from the same tank were not truly independent measures. Significant factors identified with the repeated measures ANOVA were identical to that of our univariate contrasts. Therefore, interpretations of the main effects and interactions are restricted to results of our MANOVA and univariate contrasts. Data were tested for normality and equality of variance using Kolgomorov-Smirnov and Levene's tests, respectively. Significant values for one variable were ln-transformed to minimize heteroscedasticity. All statistics were conducted with SPSS statistical software (version 13.0) at an $\alpha$ level of 0.05.

During feeding events, the 'ram-suction' index (RSI) from Norton and Brainerd (1993) was used to quantify the amount of ram versus suction used by each fish in acquiring mysid shrimp prey. This index is calculated as: RSI = $(D_{predator} - D_{prey})\ (D_{predator} + D_{prey})^{-1}$, where $D_{predator}$ is the distance moved by the predator and $D_{prey}$ is the distance moved by the prey. Pure 'ram' feeding is designated by a RSI value of 1 (only the predator moves) and pure 'suction' is defined by a RSI value of -1 (only the prey moves). Here, we assumed that values less than zero indicated suction feeding events while values greater than zero

**Table 1.** ANOVA results for variables associated with prey-capture performance in hatchery *Sciaenops ocellatus* after a 10-day rearing interval. * P ≤ 0.05 ** P ≤ 0.01

| | Mean ± SE | | |
|---|---|---|---|
| Variable | Non-vegetated | Vegetated | p-value |
| Attack Distance (mm) | 0.904 ± 0.166 | 1.309 ± 0.090 | 0.076 |
| Mean Attack Velocity (mm/sec) | 127.463 ± 24.823 | 148.163 ± 15.307 | 0.504 |
| Time to Prey-Capture (ms) | 7.722 ± 0.332 | 8.444 ± 0.544 | 0.301 |
| Maximum Gape (mm) | 3.158 ± 0.095 | 3.497 ± 0.018 | 0.013* |
| Time to Maximum Gape (ms) | 10.389 ± 0.547 | 11.278 ± 0.448 | 0.256 |
| Gape Cycle Duration (ms) | 25.278 ± 0.739 | 25.833 ± 0.500 | 0.555 |

**Table 2.** ANOVA results for variables associated with anti-predator response in hatchery *Sciaenops ocellatus* after a 10-day rearing interval. * P ≤ 0.05 ** P ≤ 0.01

| | Mean ± SE | | |
|---|---|---|---|
| Variable | Non-vegetated | Vegetated | p-value |
| Reaction Distance (mm) | 112.897 ± 12.571 | .232 ± 2.743 | 0.006* |
| 100ms Distance (mm) | 24.986 ± 5.907 | 25.573 ± 3.339 | 0.779 |
| Maximum Velocity (mm/sec) | 463.260 ± 122.227 | 639.521 ± 106.363 | 0.298 |
| Time to Maximum Velocity (ms) | 61.075 ± 6.546 | 35.347 ± 5.078 | 0.021* |
| Mean Velocity (mm/sec) | 251.348 ± 59.022 | 260.790 ± 34.274 | 0.894 |
| Maximum Acceleration ($mm/sec^2$) | 40969.314 ± 11875.029 | 74048.111 ± 19590.330 | 0.199 |

indicated ram feeding events. RSI values were used strictly for qualitative comparisons and were not compared statistically.

## RESULTS

### Prey-capture performance

Prey-capture in red drum was relatively stereotyped between habitats and prey-capture performance was not impacted by exposure to vegetation (MANOVA, $p > 0.05$). Fish reared in vegetation demonstrated slightly higher timing and magnitude of response for all variables tested (Table 1); however, univariate contrasts revealed that only maximum gape was significantly larger for fish reared in vegetated (3.497 ± 0.018 mm) versus non-vegetated tanks (3.158 ± 0.095 mm) (ANOVA, $p < 0.05$). Mean RSI values were slightly more negative for fish from vegetated tanks (-0.0947) versus non-vegetated tanks (-0.0659), indicating that individuals reared with vegetation employed a greater degree of suction during prey capture events.

### Anti-predator response

Red drum demonstrated the typical C-start escape maneuver in response to the approaching predator stimulus. Overall, anti-predator response was not found to be influenced by habitat exposure (MANOVA, $p > 0.05$); however, results of univariate contrasts revealed that reaction distance and time to maximum velocity were significantly lower for red drum reared in vegetation (ANOVA, $p < 0.01$ and $p < 0.05$, respectively). Fish reared in vegetated tanks reacted at a much shorter distance (60.232 ± 2.743 mm versus 112.897 ± 12.571 mm, Table 2) to the visual stimulus during anti-predator trials. In addition, time to maximum velocity was ~ 40% faster for red drum reared in vegetation (35.347 ± 5.078 ms) compared to fish from non-vegetated tanks (61.075 ± 6.546 ms) (Table 2). Although values for 100ms distance, maximum velocity, mean velocity, and maximum acceleration appeared to be higher for red drum reared in vegetation, no significant habitat effect was found for these variables (Table 2).

## DISCUSSION

Analysis of prey-capture performance and anti-predator response of red drum reared with and without the presence of vegetation indicated that overall survival skills (based on multivariate assessment) did not significantly improve with habitat exposure. Still, we did detect differences for a few individual variables (maximum gape, reaction distance, time to maximum velocity) which may affect the survival success of hatchery red drum. Red drum reared in vegetation demonstrated a larger maximum gape during feeding attempts, thereby allowing them to exploit a wider range of prey items (Werner 1974; Keast 1985), and possibly leading to a competitive advantage compared to individuals reared in non-vegetated tanks. In addition, these fish also possessed more negative RSI values, and therefore greater suction ability, suggesting that they may be more adept at capturing elusive prey items such as mysid shrimp. Conversely, findings from our anti-predator trials indicated that red drum may be negatively impacted by exposure to vegetation. Time to reach maximum velocity was faster for fish reared in vegetation; however, these individuals reacted at approximately half the distance to the visual stimulus during anti-predator trials compared to fish from non-vegetated tanks. Proximity to predator when response is initiated is directly related to the outcome of predator-prey encounters and larger strike distances have been associated with unsuccessful capture events (Walker et al. 2005). Therefore, red drum reared in vegetation may have less time to fully execute an escape maneuver (Dill 1974), potentially resulting in higher rates of predation-related mortality. Moreover, red drum reared in vegetation may have been less perceptive to their surroundings since these fish typically dispersed among vegetative clusters in comparison to red drum in non-vegetated tanks, which schooled in larger groups that swam more frequently.

Previous studies have demonstrated a substantial increase in survival behaviors of naïve hatchery individuals reared in more naturalistic environments (Berejikian et al. 1999; Braithwaite and Salvanes 2005; Salvanes and Braithwaite 2005). In contrast, our results suggest that habitat exposure alone may not adequately prepare red drum for the challenges faced after release. While habitat has been linked to higher survival rates during the early life stages in red drum (Rooker et al. 1998; Stunz et al. 2001; Stunz et al. 2002a), the ability to effectively utilize these areas for foraging and refuge may not be facilitated by merely exposing fish to vegetated habitat. Rather, it may be necessary to incorporate a combination of habitat and various prey types and predators in order for naïve hatchery fish to learn how to properly utilize complex habitat, thereby aiding in the development of key survival behaviors. Future studies should incorporate a suite of pre-release conditioning factors (i.e. exposure to habitat, predators, and prey types) to further clarify which factors, or combination thereof, best influence survival skills in hatchery-reared fish.

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