Evaluation of Stocking Density and Other Factors Improving Florida Largemouth Bass Fingerling Production Efficiency at the Texas Freshwater Fisheries Center Hatchery

> by Donovan Patterson

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INLAND FISHERIES DIVISION 4200 Smith School Road Austin, Texas 78744 Evaluation of Stocking Density and Other Factors Improving Florida Largemouth Bass Fingerling Production Efficiency at the Texas Freshwater Fisheries Center Hatchery

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Texas Parks and Wildlife Department Inland Fisheries Division 4200 Smith School Road Austin, Texas 78744

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ABSTRACT

The Texas Freshwater Fisheries Center Fish Hatchery (TFFC) produces Florida Largemouth Bass Micropterus salmoides floridanus (FLB) fingerlings to enhance fishing in Texas. Hatchery guidelines recommend stocking 370,657 fry/ha (150,000 fry/acre) into ponds. In 2014, TFFC began stocking approximately 321,236 fry/ha (130,000 fry/acre) to improve the average size (fish/kg) of fingerlings produced. A statistical analysis was conducted to evaluate the effect of stocking density on FLB fingerling production at TFFC. The Fish Hatchery Data System provided all the historical data for the analysis. Linear regression analysis of first cycle pond production data from 2002-2016 did not reveal a dependent relationship ($R^2 < 0.1$) between stocking density and typical measures of success (e.g. percent survival, harvest density, growth, average length, and average weight). The data exhibited significant variability although ponds stocked at lower densities tended to be more consistent. The TFFC has only attempted two target stocking densities, ~370,657 fry/ha (~150,000 fry/acre) and ~321,236 fry/ha (~130,000 fry/acre) over the period studied, and occasionally ponds were stocked below target densities (these low-density ponds were generally stocked at ~284.170 fry/ha or ~115,000 fry/acre) out of necessity. Categorical comparison (ANOVA) of the two target densities (High: ~370,657 fry/ha or ~150,000 fry/acre and Medium: ~321,236 fry/ha or ~130,000 fry/acre) and the lower density (Low: ~284,170 fry/ha or ~115,000 fry/acre) did not reveal a significant difference in the average harvest densities of the three treatments (P=0.1151). However, percent survival and fish/kg were significantly improved when ponds were stocked at ~321,236 fry/ha $(\sim 130,000 \text{ fry/acre})$ versus $\sim 370,657 \text{ fry/ha}$ ($\sim 150,000/acre)$ (P < 0.0001). Neither of these treatments was statistically dissimilar when compared to stocking ~284,170 fry/ha (~115,000 fry/acre). Measures of success were highly variable for all treatments suggesting that slight deviation in stocking density was independent of FLB fingerling production success. Using the same data and incorporating zooplankton densities and water quality data; a multivariate analysis was performed to determine what variables were most correlated to FLB fingerling production measures of success and to attempt to isolate sources of variability. Correlation coefficients associated between time (Year) and other variables were within the range $(0.3 < |\mathbf{R}| < 0.5)$ suggesting that operational decision making has improved over time. No other strong correlations were observed (R < 0.3). We recommend continuing to target a stocking density of ~321,236 fry/ha (~130,000 fry/acre) at TFFC as this strategy poses significant operational advantages and does not appear to negatively impact production.

INTRODUCTION

Texas Parks and Wildlife Department (TPWD) hatcheries raise several popular sport fish species and stock them into public waters to maintain and promote fishing opportunities in Texas. The most popular species is the Florida Largemouth Bass *Micropterus salmoides floridanus* (FLB). The methodology used by TPWD hatcheries to culture FLB is published in Glenewinkel et al. (2011). Black bass culture typically follows three phases: spawning, hatching, and grow-out. To commence the grow-out phase, fish fry are stocked into fertilized hatchery ponds ~6–10 days post-hatch. Glenewinkel et al. (2011) recommends stocking fry at a density of 370,657 fry/hectare (150,000 fry/acre) to consistently meet the size and production goals (38-mm average length and 1,450 fish/kg average).

The Texas Freshwater Fisheries Center Fish Hatchery (TFFC) is one of the primary fish hatcheries used to produce FLB. The TFFC draws water from Lake Athens, a small municipal impoundment. Lake Athens is an oligotrophic water source and has low hardness (~ 28 mg/L) and alkalinity (~ 17 mg/L) levels. Florida Largemouth Bass fry respond poorly to artificial diets and instead must rely on live prey, specifically zooplankton and insect larvae, in culture ponds (Keast and Eadie 1985; Colgan et al. 1986). Zooplankton species (especially Copepods and Cladocera) require calcium to form their exoskeletons (Wærvågen et al. 2002; Arnott et al. 2017; Azan and Arnott 2018). The low calcium and chloride concentrations in Lake Athens limit zooplankton population density and microorganism size in the hatchery ponds at TFFC when compared to those found at the other TPWD hatcheries with harder and more productive water. As a result, TFFC has struggled to consistently meet the production goals of 38-mm average length and ~1,450 fish/kg.

Aquaculturists commonly reduce stocking density to increase growth rate and harvest size in the same time frame (Piper et al. 1982). Most stocking density literature focuses on production of market-size fish for human consumption. Teng et al. (1978) found that increasing stocking densities from 60 to 120 fish/m³ in net pens decreased weight gain, feed efficiency, mean fish weight, and survival of Estuary Grouper *Epinephelus salmoides*. Similarly, Dambo (1993) found that mean length, weight, and specific growth of Nile Tilapia *Oreochromis niloticus* were reduced when fry were stocked at higher densities. Castillo-Vargasmachuca et al. (2012) reported that the mean individual weight of Pacific Red Snapper *Lutjanus peru* reared in floating net pens was inversely related to stocking density and that mean final weight, weight gain, and specific growth rate were highest for fish stocked at the lowest density. The mean weight of African Catfish *Clarias gariepinus* reared in cages was highest at lower densities (Hengsawat et al. 1997). Growth and survival of Sea Bream *Archosargus rhomboidalis* was reduced by increased stocking density in laboratory conditions (Houde 1975).

Limited research has been published concerning stocking density of carnivorous fish fry in freshwater ponds for stock enhancement purposes. The large number of confounding site-specific variables resulting in highly variable data is likely a factor excluding such research from publication. Increasing the stocking density of Walleye *Stizostedion vitreum* from 250,000 to 375,000 fry/ha increased the gross yield without impacting total percent survival (Harding and

Summerfelt 1994). However, the average size of the fish harvested was smaller in ponds stocked at higher densities. Flowers and Fox (2011) reported that mean length and weight of Walleye at harvest showed an inverse logarithmic relationship with initial stocking density, whereas biomass production was positively related. Culver et al. (2010) reported that increasing the stocking density of Yellow Perch *Perca flavescens* cultured in ponds from 150,000 to 300,000 fry/ha increased the total yield from 45 to 100 kg of fish/ha. Kurten (2001) investigated the effects of various pond variables (including stocking density) on FLB fingerling production at the now retired TPWD Jasper State Fish Hatchery in earthen ponds. He reported that stocking fry at 19 mm TL at a density of ~400,000 fry/ha (~161,875 fry/acre) or less maximized survival.

In 2014, TFFC deviated from the production manual recommendations and began stocking ~321,236 FLB fry/ha (~130,000 FLB fry/acre) to attempt to meet the target average size of 38 mm. However, lowering the stocking density can possibly reduce production efficiency in terms of numbers harvested. To address this concern, a statistical comparison of the effect of stocking density on production measures of success was conducted.

METHODS

Data collection

Historical data (2002–2016) was obtained from the Fish Hatchery Data System (FHDS) where TPWD compiles fish production data. Additional data, collected from the FHDS for multivariate analysis, is shown in Table 1.

Only data from the first cycle of production each year was compiled because fry availability, not stocking density, is the limiting factor for second cycle production. Production data was compiled using Microsoft Excel and filtered to exclude: ponds with incomplete data, ponds stocked with advanced fry (> 7 mm), ponds that experienced partial and split harvests, and ponds that experienced 0% survival (since stocking density could not have been the limiting factor in those instances).

Statistical analysis

The statistical analysis software JMP-12 (SAS Institute, Cary, North Carolina) was used for statistical analysis. Linear regression analysis was performed on the compiled data set using stocking density (fish/acre) as the independent explanatory variable and harvest density (fish/acre), total harvest weight (kg/acre), survival (%), growth (mm/day), average fish length (mm), and average fish size (fish/kg) as the dependent variables. All data was retrieved from the FHDS on a per pond basis. Variables were considered to have a linear relationship if the R² value for trend line fit was > 0.5.

Linear regression analysis highlighted that two distinct stocking density ranges had been most used by TFFC (308,881-358,302 fry/ha [125,000-145,000 fry/acre] and 358,302-395,367 fry/ha [145,000-160,000 fry/acre]), but the lack of available fry occasionally resulted in a pond being stocked at a lower (non-target) stocking density. Since linear regression did not suggest a relationship between success and stocking density, a categorical comparison of high, medium, and low stocking density ranges was performed to evaluate effects of stocking density on measures of production success. Stocking density categories were defined as > 358,302 fry/ha

(145,000 fry/acre) as "high density", 308,881–358,302 fry/ha (125,000–145,000 fry/acre) as "medium density", and 259,460-308,881 fry/ha (105,000–125,000 fry/acre) as "low density". Ponds stocked at a density less than 259,460 fry/ha (105,000 fry/acre) were excluded from this comparison since stocking below this level is only done out of necessity in very rare circumstances. An ANOVA was used to determine significant differences among stocking density categories with regards to harvest density (fish/acre), total harvest weight (kg/acre), percent survival, growth (mm/d), average fish length (mm), and average fish size (fish/kg). Differences were considered significant if P < 0.05. A Tukey's HSD test was conducted to specify any significant differences.

RESULTS

Linear regression analysis

No linear relationship was observed between stocking density and harvest density (Figure 1), total harvest weight (Figure 2), survival (Figure 3), growth (Figure 4), average fish length (Figure 5), or average fish weight in fish/kg (Figure 6).

R² values for all dependent variables did not approach 0.5, suggesting that a decrease in stocking density to ~321,236 fry/ha (~130,000 fry/acre) in 2014 was not primarily responsible for changes in production success at TFFC between 2002 and 2016. There also appear to be two distinct target stocking density ranges and range below target densities that could possibly be compared categorically.

Categorical comparison of stocking densities

Harvest density did not differ significantly between stocking density categories. Medium stocking density ponds exhibited significantly higher survival, faster growth, and significantly lower fish/kg ("higher quality fish") than high density ponds; however, neither differed significantly from the low stocking density category (Table 2). The medium stocking density range also produced the longest average fish length and significantly more kg/acre than either the high or the low stocking density ranges. High variability is partly the result of categories possessing very different sample sizes (n) (Table 2).

A categorical comparison revealed a more significant relationship between stocking density and production success but there was still a high amount of variability that stocking density alone could not explain. A question that became apparent during the analysis was "What variables most correlate with production success?" A multivariate analysis was completed comparing all the variables listed in Table 1. Variables were considered to be correlated if |R| > 0.5; where R represents the correlation coefficient.

Post hoc multivariate analysis

The only relationship observed that was correlated (|R| > 0.5) was between Year and Harvest weight (kg/acre, R = 0.5159). Several other correlation coefficients between Year and other measures of success showed weaker but nearly relevant relationships (0.3 < |R| < 0.5, Table 3). While several significant relationships between stocking density and success were observed, all were weak correlations (Table 4). No other significant correlation was observed between the variables listed in Table 1.

DISCUSSION

The lack of a linear relationship ($\mathbb{R}^2 < 0.5$) between stocking density and various measures of production success suggests that stocking density is not the determining factor of production success during the first cycle of FLB production at TFFC. However, it is evident in Figures 1–6 that TFFC has targeted only two stocking densities: ~321,326 fry/ha (~130,000 fry/acre) and ~370,657 fry/ha (~150,000 fry/acre). The relationship between stocking density and success would likely be more evident had more target stocking densities been attempted. Finding the high and low densities at which stocking density begins to significantly affect production success would require TFFC to stock ponds at a variety of densities (both above and below historical target densities) which could negatively impact FLB production.

A categorical comparison of continuous data such as stocking density has inherent weaknesses. However, it is appropriate to use in this situation as TFFC has only targeted two stocking density ranges and most of the data analyzed does not deviate greatly within those ranges (Table 2). While data exhibit high variability, harvest densities did not differ significantly for fry stocked at a rate of between 259,460 fry/ha (105,000 fry/acre) to 395,367 fry/ha (160,000 fry/acre) (Table 2). There was no evidence that lowering the target stocking density at TFFC in 2014 to 321,236 fry/ha (130,000 fry/acre) did not impact gross yearly production efficiency. In fact, the "medium" stocking density range exibited higher fish survival, higher average weight (fish/kg) and average length (mm), higher total biomass harvested (kg/acre), and faster growth (mm/day) than the "high" density, suggesting that it is more efficient and beneficial to individual fish condition to stock at ~312,236 fry/ha (~130,000 fry/acre) versus ~370,657 fry/ha (~150,000 fry/acre). This has reduced the first cycle fry requirement at TFFC, allowing the second cycle to commence earlier each year. Decreasing the target stocking density further (i.e., stocking at a "low" density, ~284,170 fry/ha (~115,000 fry/acre) did not significantly change fish survival, growth (mm/day) or average weight (fish/kg) compared to the "medium" and "high" target stocking densities, although sample size for the low density category was limited.

The high variability observed is partly due to the varying sample sizes between the stocking density categories. As suggested by the linear regression analysis, high variability could also indicate that, while target stocking densities and production success may be correlated when compared categorically; stocking density was not the direct cause of the success. In fact, another variable that changed in the same time frame as the decrease in stocking density may be directly or indirectly responsible for the difference in production success exhibited by the categorical comparison of stocking densities.

The multivariate analysis suggested that production has become more consistent over time. Correlation coefficients associated with the time variable "Year" and various measures of success were the most correlated values observed. Harvest weight, average harvest length, kg/acre harvested, and growth (mm/day) showed slight positive correlations with time, and stocking density and fish/kg showed slight negative correlations with time (Table 3). These correlations indicate that over the past 14 years, decisions to reduce stocking rates at TFFC have resulted in moderate improvements to production efficiency of FLB.

Some correlation coefficients between stocking density and other variables hinted at weak correlations. Gross harvest weight, average harvest length, kg/acre harvested, and harvest density were all negatively correlated with stocking density; meaning these measures of success are negatively impacted by increasing the stocking density (Table 4). The only positively correlated variable with stocking density that had R value approaching 0.5 is the number of fish/kg at harvest. Positive correlation in this case suggests that stocking at a higher density would result in smaller fish (i.e. an increase in number of fish/kg) (Table 4). This is congruent with the categorical comparison that indicated fish produced at lower stocking densities were more robust (in terms of number of fish/kg) than fish produced at a higher stocking density (Table 2).

Other variables showed weak correlative relationships ($|\mathbf{R}| < 0.4$). Many of these variables are outside of the control of the hatchery staff (e.g., temperature and DO). These variables likely contributed to the high variability observed when trying to compare the effect of stocking density on production success.

Management implications

The decrease in stocking density implemented by TFFC had a positive effect if any. Statistical observations indicate that a larger or at least equal number of more robust fish can be produced by stocking ponds at a lighter density without impacting harvest density. The large number of dependent and independent variables, both measured and unmeasured, make any analysis difficult. Measuring one variable independent of another is impossible (without being highly intentional), particularly when using production data. It is entirely possible that the optimum stocking density is not a single number but a range and that all attempted stocking densities could already fall within that range (possibly explaining low levels of significance with high levels of variability).

The most influential factor on production success at TFFC has likely been time. Staff at TFFC have observed patterns and pitfalls associated with production of FLB fingerlings at TFFC over many years and have adjusted their decision making to optimize production. One such operational decision was to stock fish at a slightly lower pond stocking density because it offers several operational advantages such as completing the first cycle of pond production faster leaving more time during optimal weather conditions to begin and complete the second cycle of pond production.

RECOMMENDATIONS

- Continue to stock FLB fry rearing ponds at TFFC at a density of ~321,236 fry/ha (130,000 fry/ acre)
- If the relationship between stocking density and production success is to be further investigated, TFFC should be more intentional about stocking at densities much lower and much higher than previously attempted to establish the points at which stocking density begins to affect production success.

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TABLE 1. Variables compared using multivariate analysis.

Production Variables	Zooplankton Variables	Water Quality Variables
Location	Mean Cladocera/L	Average AM DO (mg/L)
Pond Size (acres)	Mean Copepod Adult/L	Average PM DO (mg/L)
Year	Mean Copepod Nauplii/L	Average AM pH
Stock Date	Mean Rotifer/L	Average PM pH
Harvest Date	Mean Total Zooplankton/L	Average AM Temp (°C)
Culture Period (days)	Median Cladocera/L	Average PM Temp (°C)
Stocking Density (fish/acre)	Median Copepod Adult/L	Maximum AM DO (mg/L)
Harvest Density (fish/acre)	Median Copepod Nauplii/L	Maximum PM DO (mg/L)
Gross Harvest Weight (kg)	Median Rotifer/L	Maximum AM pH
Fish/kg at Harvest	Median Total Zooplankton/L	Maximum PM pH
Avg. Fish Length at Harvest (mm)	Maximum Cladocera/L	Maximum AM Temp (°C)
Growth (mm/day)	Maximum Copepod Adult/L	Maximum PM Temp (°C)
Survival (%)	Maximum Copepod Nauplii/L	Minimum AM DO (mg/L)
	Maximum Rotifer/L	Minimum PM DO (mg/L)
	Maximum Total Zooplankton/L	Minimum AM pH
	Minimum Cladocera/L	Minimum PM pH
	Minimum Copepod Adult/L	Minimum AM Temp (°C)
	Minimum Copepod Nauplii/L	Minimum PM Temp (°C)
	Minimum Rotifer/L	
	Minimum Total Zooplankton/L	













Table 2: Means and standard deviations of categorical comparison of stocking density and measures of success

Density Range	High density (n=195)	Medium density (n=80)	Low density (n=19)	Р
Average stocking density (fry/acre)	151,867±2,318	128,402±4,560	117,686±6,534	
Average harvest density (fish/acre)	99,319±30,044	99,016±24,786	85,061±28,962	0.1151
Survival (%)	$65 \pm 20 \text{ b}$	77 ± 20 a	$72 \pm 20 ab$	< 0.0001
Average fish weight (fish/kg)	$1,\!848\pm638~\mathrm{b}$	$1,352 \pm 560 \text{ a}$	$1,568 \pm 401 \text{ ab}$	< 0.0001
Growth (mm/day)	$1.05\pm0.18~b$	1.16 ± 0.16 a	1.12 ± 0.17 ab	< 0.0001
Average Length (mm)	$40.1\pm2.9~\text{b}$	$43.3 \pm 4.0 \text{ a}$	$40.9\pm2.7~b$	< 0.0001
Average Biomass (kg/ acre)	$56.7\pm19.4~b$	80.5 ± 29.6 a	$56.8 \pm 25.5 \text{ b}$	< 0.0001

High density: > 145,000 fry/acre, Medium density: 125,000–145,000 fry/acre, Low density: 105,000–125,000 fry/acre

Production variables		Correlation coefficient	Probability
Year	Gross Harvest Weight (kg)	0.4187	< 0.0001
Year	Avg. Fish Length at Harvest (mm)	0.4268	< 0.0001
Year	kg/acre Harvested	0.5159	< 0.0001
Year	Stocking Density (fish/acre)	-0.3960	< 0.0001
Year	Growth (mm/day)	0.3815	< 0.0001
Year	Fish/kg at harvest	-0.3594	< 0.0001

TABLE 3. Correlation coefficients between year and select measures of success. (n = 294).

Produ	ction variables	Correlation coefficient	Probability
Stocking Density	Gross Harvest Weight (kg)	-0.3031	< 0.0001
Stocking Density	Avg. Harvest Length (mm)	-0.3810	< 0.0001
Stocking Density	kg /Acre Harvested	-0.3336	< 0.0001
Stocking Density	Harvest Density (fish/acre)	0.0512	0.4197
Stocking Density	Fish/kg at harvest	0.3457	< 0.0001

TABLE 4. Correlation coefficients between stocking density and select measures of success. (n = 294).

Texas Parks and Wildlife Department 4200 Smith School Road, Austin, Texas 78744

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