Effect of Increased Egg Stocking Density in Existing and Experimental Catfish Incubators

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Abstract: Channel catfish egg masses are typically incubated in stationary wire mesh baskets suspended in metal troughs with flow-through water that is agitated and circulated between the baskets and around the eggs with rotating paddles. A limiting factor in the successful incubation of channel catfish egg masses is the absorption of oxygen by the developing embryos; low levels of dissolved oxygen in the water result in premature hatching and increased fry mortality. We modified and tested a vertical-lift incubator (the "See-Saw") for incubating channel catfish egg masses. Both the See-Saw and control (paddle-type) troughs were loaded with 26 egg masses (13.2 kg) per trough which is 1.5–2.0 times higher than recommended loading rates. Swim-up fry survival was 2.3-fold higher for the See-Saw than the control troughs. Many of the sac-fry produced in the control troughs were either dead when removed or died prior to reaching swim-up stage, presumably due to oxygen stress. The See-Saw incubator increases fry survival while simultaneously using less water and hatchery space per unit fry produced.

Key words: catfish, incubator, See-Saw, oxygen

Incubators for hatching channel catfish (*Ictalurus punctatus*) eggs have used a design nearly unchanged for the past 80 years (Clapp 1929). Incubators typically consist of paired metal troughs with multiple paddles rotating on a metal shaft above each trough (Avery and Steeby, 2004). Catfish egg masses (10,000–20,000 eggs attached together in a gelatinous matrix) are placed in 0.64 cm mesh hardware cloth baskets suspended in the water between paddles in the trough. The rotating paddles are designed to circulate water through and among egg masses, simulating the fanning provided by the male catfish (Tucker and Robinson 1990). Depending on the size of the trough, typically 7 to 9 kg of eggs are incubated per trough. Egg masses incubated at 26 C hatch in six days (Small and Bates 2001) and are transferred to fry-rearing tanks before being stocked in ponds.

The main factor limiting hatch rate appears to be the dissolved oxygen (DO) concentration around developing embryos (Torrans and Steeby 2008). Low DO concentration not only increases mortality, but also can lead to premature hatching. Oxygen demand increases as the number of eggs stocked in a trough increases, limiting the amount of eggs that can be placed in a trough. Increasing water flow can raise the amount of oxygen supplied, but may not always be practical due to either the high cost of heating the water or a limited supply of usable water (Tucker 1991, Avery and Proc. Annu. Conf. Southeast. Assoc. Fish and Wildl. Agencies 54:131-135

Steeby 2004). A higher DO concentration can be achieved with the addition of liquid oxygen or atmospheric air, either by aerating the main water supply or through airstones in individual troughs. However, as the mass of spawns in a hatching trough increases, water circulation around and through individual egg masses is reduced, decreasing the amount of oxygen delivered to the center of each egg mass, causing death of embryos in the center (Avery and Steeby 2004). This may lead to subsequent fungal or bacterial infections affecting the remainder of the egg mass (Tucker 1991).

Torrans et al. (2009) introduced a new catfish egg mass incubator called the "See-Saw," a vertical lift incubator that moves the egg masses in and out of the water. The See-Saw was designed to improve water movement through the center of the egg mass and circulation around individual spawns. Furthermore, Torrans et al. (2009) believed that the eggs consume atmospheric oxygen while out of the water, thus reducing demand for oxygen in the water. Compared to a standard hatching trough, the See-Saw increased fry production and decreased the amount of water used per fry produced.

We designed this study to compare the ability of the See-Saw and traditional hatching troughs to produce catfish fry at an egg density higher than recommended levels. To quantify the effects of high loading rates we measured hatch rate, survival to swim-up, and selected water quality variables (temp, DO, flow, pH, TAN, and NO_2 -N) for both incubator types. These measurements provide an assessment of potential advantages that the See-Saw may possess over standard hatching troughs.

Methods

Study Site

This project was completed at Needmore Fisheries LLC, Glen Allen, Mississippi, a commercial hatchery in western Mississippi. All channel catfish eggs were collected from spawning ponds in the usual manner and hatched on site by farm employees during June 2009.

Incubator Design

Torrans et al. (2009) detailed the construction and mechanics of the See-Saw incubator. The movement of the incubator is similar to that of a beam balance (or see-saw) in that a horizontal bar pivoted in the middle lifts the rack (containing three baskets) in one trough up and out of the water while the rack in the adjacent trough is lowered in to the water. The incubator consisted, briefly, of an angle-iron frame bolted to two pairs of existing hatching troughs, creating a unit that operated four troughs (Figure 1). A single phase motor oscillated at 6 cycles per minute, rotating a solid steel shaft that runs the length of the incubator. The steel shaft had four crossbars welded to it that extend equidistant over each side of the incubator. The crossbars were connected by chain to rectangular racks constructed of 5-cm angle-aluminum that support the hatching baskets. Hatching baskets were constructed of 0.64-cm square mesh PVC-coated hardware cloth, were approximately 61 x 43 x 8 cm, and contained a cross-partition in the center to evenly distribute the egg masses. The baskets had a hinged lid and a 2.54-cm lip to contain the eggs. Three baskets were secured to each rack using a bolt, washer, and wing nut through the center of each basket. This bolt system also served to keep the lid closed, as did bungee cords strapped to both the basket and rack.

Egg and Fry Sampling, Stocking, and Harvest

Eggs, sac fry, and swim-up fry densities were calculated using sample counts at each stage. Eighteen egg samples from spawns from both treatments were weighed $(7.62 \pm 0.43 \text{ g/sample};$ mean ± SE) and counted to determine the number of eggs/kg and to estimate the number of eggs per trough (Torrans and Lowell 2001). Sample counts of eggs during this experiment averaged 35,853 ± 828 eggs/kg. Sac fry were sampled the morning after hatching by netting and measuring fry from a hatching trough in a graduated cylinder ($4.6 \pm 0.3 \text{ ml}$ of fry per sample) and counting the fish in each sample. Counts of sac fry averaged $53.4 \pm 1.3 \text{ fry}/$



Figure 1. Photograph of a See-Saw incubator used in this study (prior to loading eggs). Note that as the left rack is up in the air, the right rack is down in the water. Each rack contains three hatching baskets that are secured to the rack. The water supply for these two troughs is in the foreground and the drain is at the far end in each trough. The following components are labeled: (A) 6 rpm motor, (B) hatching trough, (C) angle-aluminum frame supporting the See-Saw, (D) steel shaft running the length of the troughs, (E) crossbars, (F) angle-aluminum rack that holds the baskets, (G) hatching baskets, and (H) oxygen supply (not used in this study).

ml. Swim-up fry sample counts were measured the day of swimup by netting samples of fry $(3.69 \pm 0.14 \text{ g of fry per sample})$ from troughs. Samples were patted dry, weighed, and counted, averaging $37.5 \pm 1.3 \text{ fry/g}$.

The typical loading rate of troughs for this particular hatchery is approximately 12 spawns per trough, which is similar to the recommended numbers for the industry (Avery and Steeby 2004). We loaded both control troughs (standard hatching troughs) and experimental troughs (See-Saws) with the same number (26) of 1-to 3-day old randomly-selected spawns per individual trough, approximately double that of the recommended rates.

Sac fry were siphoned from the incubators as they hatched (typically over the course of one to two days), sampled and measured volumetrically in a 1000 mL graduated cylinder to determine the total number removed per trough, and then transferred to a rearing trough. As fry reached the swim-up stage (four to five days later), they were sampled and weighed to determine total swim-up fry production.

Water Quality

We measured DO (mg/L), pH, temperature (C), and water flow (L/min) daily in both control and See-Saw troughs. Water flow measurements were taken from the inlet pipe. Temperature, DO, pH, total ammonia nitrogen (TAN; mg/L), and nitrite-nitrogen concentration (NO₂-N; mg/L) were measured at the head (inlet end) of the trough and at the drain. Dissolved oxygen, pH, and temperature were measured using the Hach HQ40d Multi-Parameter Digital Meter (Hach Company, Loveland, Colorado). Total ammonia nitrogen and nitrite concentration were measured using the Hach DR 2800 spectrophotometer and Hach TAN and Nitrite assays (Hach Company, Loveland, Colorado). Because inlet values of these parameters varied over time and originated from one or more of four separate wells, we calculated the change from the water inlet to drain in individual troughs to serve as indicators of oxygen consumption and ammonia and nitrite production. Water entered each trough from an inlet pipe and flowed through once, leaving from an overflow pipe. Water flow did not differ between treatments, averaging 9.0 ± 0.5 and 10.3 ± 0.6 L/min for the control and experimental troughs, respectively.

Statistical Analyses

Means of individual troughs in both treatments were compared for fry production, fry removal, and water quality were determined using either a pooled or Satterthwaite two sample *t*-tests for populations of equal or unequal variances, respectively. SAS Enterprise Guide 4.2 (SAS Institute, Inc. 2008) was used in all analyses. We designated the level of significance at P<0.05.

Results

Hatch Rates and Production

We incubated the same number and mass of spawns and number of eggs in both the control and See-Saw troughs (Table 1). Although both incubators produced similar numbers of sac fry at

Table 1. Water	guality, stocking,	and production	parameters from contro	I and See-Saw troughs.
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Variable	Control	See-Saw	Р
n	4	4	
Temp (C)	25.9 ± 0.3	26.4 ± 0.2	0.237
Flow (L/min)	9.0 ± 0.5	10.3 ± 0.6	0.125
Spawns (n)	25.8 ± 0.5	25.5 ± 1.0	0.823
Mass (kg)	13.16 ± 0.11	13.27 ± 0.08	0.457
Eggs (n)	472,878 ± 3,892	474,947 ± 2,924	0.689
Hatch time (days)	5	6	
Sac fry moved (n)	306,716±32,575	303,179±33,304	0.941
Egg to hatch survival (%)	64.8±6.7	64.0 ± 7.5	0.938
Swim-up fry produced (n)	109,525±30,324	253,734 ± 31,935	0.017
Egg to swim-up survival (%)	23.3 ± 6.5	53.6±7.1	0.020



Figure 2. Mean daily change in D0 concentration (A), daily number of sac fry moved from incubators (B), daily change in TAN (C), and daily change in nitrite-nitrogen (D). Water quality parameters were calculated as the change from the inlet to the drain of See-Saw and control troughs. Water quality parameters measured were largely the same for both treatments but were not measured for control troughs on day six as all of the eggs were hatched by that point.

hatch, eggs in the control troughs hatched a full day earlier than eggs in the See-Saw troughs. Two- and 1.5-fold more sac fry were removed from the control troughs than the See-Saw on days three and four, respectively, while 2.6- and 6.5-fold more fry were moved from the See-Saw on days five and six, respectively (Figure 2). The See-Saw incubator produced a significantly higher number of swim-up fry than the control troughs, which yielded a 2.3-fold increase in survival to swim-up (Table 1).

Water Quality

Water entering the control and See-Saw troughs had the same mean DO concentration over the entire incubation duration, averaging 7.88 ± 0.08 and 7.84 ± 0.09 mg/L ($97.0 \pm 1.0\%$ and $97.3 \pm 0.9\%$ saturation), respectively. Dissolved oxygen concentration of water leaving the trough did not differ between treatments and the change from the head to end of trough (0.4-1.1 mg/L) was not different between treatments on any day (Figure 2). Inlet pH was the same (8.23 ± 0.03) for both treatments over the incubation period as well as the pH at the drain, which averaged 8.20 ± 0.01 and 8.22 ± 0.01 for the control and See-Saw troughs, respectively. Total

ammonia nitrogen and nitrite production remained largely unchanged for both treatments throughout incubation, although the control troughs had higher TAN production on day one (Figure 2).

Discussion

At an increased loading rate in identical troughs, the See-Saw incubator produced more fry than a traditional paddlewheel incubator with the same water flow. Lifting the eggs through and out of the water apparently provides physiological benefits to the developing embryos as well as production benefits to the farmer. We discuss the potential underlying reasons for the success of the See-Saw incubator below.

Our results indicate that the control troughs produced similar numbers of total sac fry as the See-Saw troughs. However, many of the sac fry in the control troughs were visibly dead in the control incubators, resulting in the large increase in TAN production in the control troughs on day four (Figure 2). When we siphoned from the control incubators we were unable to separate the live from the dead fry; therefore they were measured and calculated together for hatch rate. This accounts for the high hatch rate recorded yet low survival to swim-up for the control troughs (Table 1). Reported hatch rates for most commercial production facilities average 60% (Avery and Steeby 2004). The See-Saw hatch rate ($64.0 \pm 7.5\%$) and survival to swim-up ($53.6 \pm 7.1\%$) fall in line with the industry average.

When incubating at the measured temperature (26 C), spawns typically hatch in 6.25 days (Small and Bates 2001). We began removing sac fry on day three of incubation and finished on day six, which reflects the age range of egg masses when we stocked them. However, eggs in the control troughs largely finished hatching on day five, one day earlier than See-Saw troughs and published averages. Torrans and Steeby (2008) demonstrated that eggs will hatch prematurely in sub-optimal oxygen conditions, which leads to increased mortality and deformities, suggesting that the control treatment experienced low DO concentration in the center of the spawns.

Measured water quality parameters were predominantly the same for both treatments. We expected to find lower oxygen consumption in the See-Saw troughs; however, the change in DO concentration was statistically the same for both the See-Saw and control troughs. On average, the See-Saw troughs had lower oxygen consumption than the control troughs on day two, the same on day three, and higher on days four and five. These differences may be reflective of the superior ability of the See-Saw to aerate eggs on day two and the faster removal of fry from the control troughs on days three and four (Figure 2). Despite a comparable profile in water quality, the See-Saw produced more than twice as many swim-up fry than the standard troughs when stocked at double the normal suggested density (Table 1). Given that the change in DO concentration was the same for both treatments, we believe that the crowding of spawns in the paddlewheel treatment reduced mixing of water among the spawns and reduced oxygen transfer to the center of spawns, resulting in lower fry survival in those spawns or areas of spawns.

The See-Saw's unique incubating movement of pulling and pushing the spawns through the water potentially provided better aeration to the center of spawns and prevented spawns from suffocating one another when loaded at high densities. Even at high densities, spawns in the See-Saw separated and repositioned in the basket with each up and down stroke, likely preventing any space- and density-generated aeration problems. Furthermore, it is plausible that the eggs were absorbing oxygen directly from the atmosphere while they were in the air, effectively reducing the demand for oxygen placed on the water. Finally, the action of raising the racks out of and then splashing back into the water certainly accounted for an increase in water aeration. A combination of all these mechanisms likely accounted for the See-Saw's ability to successfully incubate an increased number of eggs using the same area and water volume as traditional paddlewheel incubators.

The See-Saw may enable commercial or public sector hatcheries to incubate the same number of channel catfish fry in half or less the space or less than previously used, using the same or less volume of water or less. A previous project with the See-Saw (Torrans et al. 2009) indicated that 200% – 400% more eggs may be incubated in the same area as used by existing hatching techniques, enabling a hatchery to hatch more catfish in the same space or devote more space to other fish species. Additionally, an incubator using two- to four-fold less water decreases pumping and heating costs.

Future modifications of the See-Saw may be needed; however, we conclude that the concept per se of lifting the eggs through and out of the water appears to be beneficial. If the eggs are indeed consuming oxygen while out of the water, it may be advantageous to increase the duration of time that they are out of the water. Planned studies will measure oxygen concentration inside spawns that are incubating using both the traditional and See-Saw incubators. Additionally, the amount of water previously thought needed to successfully incubate eggs may be decreased even further than in the current study. Further studies into the dynamics of the See-Saw could demonstrate how many eggs or how little water is possible with the incubator.

Acknowledgments

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