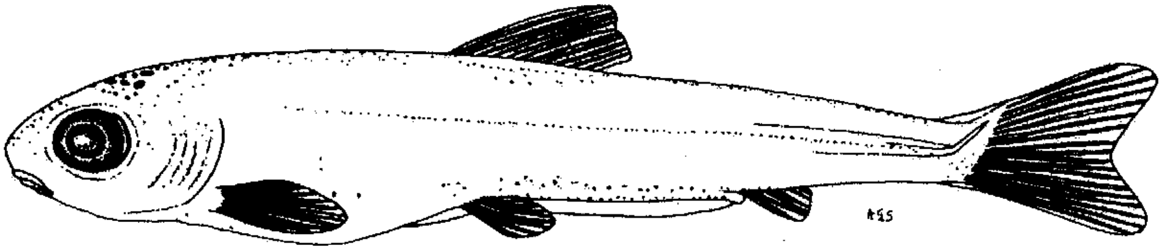
Culture Techniques and Ecological Studies of the Robust Redhorse *Moxostoma robustum*: assessment of reproductive and recruitment success, and percent fine sediment on hatching success

1998 Annual Report



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Task 1. Artificial Propagation of Robust Redhorse (Moxostoma robustum)

Broodfish Collection

Artificial propagation of robust redhorse for research and husbandry purposes continued this year much as in previous years. Adult robust redhorse were collected from the Oconee River by Georgia DNR biologists during May 18-22, 1998. The objectives of this effort were to provide sufficient numbers of male and female robust redhorse in spawning condition to produce healthy fertilized embryos for: 1) hatchery rearing of robust redhorse to meet stocking goals established by the Robust Redhorse Conservation Committee, 2) experimental studies approved by the Robust Redhorse Conservation Committee, and 3) improving the efficacy of current propagation techniques. Low flows from Sinclair Dam were provided by Georgia Power Company, and broodfish were captured by electrofishing on May 18-22, 1998. Fish were transported to a temporary spawning facility constructed on the river bank next to the Beaverdam Wildlife Management Area boat ramp (refer to Robust Redhorse Conservation Committee Annual Report, 1997 for details of broodfish collection). A total of 32 specimens were collected during this period (17 males and 15 females). Fourteen (14) females and 17 males were transported to the spawning facility. Fish were anesthetized (10 ppm Metomidate) prior to handling. Fin clips were collected and preserved in ethanol for subsequent genetic evaluation. PIT (Passive Integrated Transponder) tags were inserted in all fish. Lengths (mm TL) and weights (Kg) were recorded for all fish, and ovulation stage of females was determined.

Spawning

Of the 14 females received at the Beaverdam spawning facility, 5 fish were either found to be overripe at the time of initial examination, or subsequently released overripe eggs and ovarian fluid after hormone-induced ovulation. Three (3) other females did not exhibit indications of progression towards ovulation, even after hormone injection. One female was received at the spawning facility with several external lesions and no effort was made to spawn this fish. Ripe eggs were collected from 4 females, two of these fish were spawned on two consecutive days, the other two were spawned on three consecutive days. There were two mortalities during this period, all other fish were released at the Beaverdam WMA boat landing. Fertilized eggs were water hardened and provided to investigators for use in laboratory studies or were transported in insulated fish-shipping boxes by personnel from the Piedmont National Wildlife Refuge to three incubation facilities. Table 1.1 provides basic information on fish received at the spawning facility. Table 1.2 provides information on matings and embryo production. Based on observations from previous years, the peak of spawning activity is expected to occur in the first and second weeks of May. Spawning activity in the spring of 1998 appeared to be delayed somewhat, probably by heavy rains during the winter and early spring. However, by May 22, 1998, water temperatures in the Oconee river had risen above 25°C and spawning activity appeared to end abruptly.

Table 1.1. Information on Floy and PIT tags, total length in mm (TL), weight in grams (WT), and capture date for adult robust redhorse received at Beaverdam WMA Spawning Facility for spawning during spring of 1998.

Floy Tag	Floy Tag	PIT Tag	TL	WT	Captured	Sex
00409	00410	4033715841	667	4900	5/18/1998	female
00585	00600	4034077D45	708	6130	5/19/1998	female
00563	00591	4033632412	653	6750	5/19/1998	female
00123	00730	4033745631	540	2900	5/20/1998	female
00570	00571	4033792B05	640	4210	5/18/1998	male
00489	00490	403400620F	641	4400	5/18/1998	male
00066	00450	4033767158	612	3750	5/19/1998	male
00539	00561	40332E5A5D	642	3750	5/19/1998	male
00092	00093	40337F2506	603	3300	5/19/1998	male
00559	00587	40337F795D	657	4200	5/19/1998	male
00142	00560	40335E7736	654	4500	5/19/1998	male
00301	00586	40336A2C17	628	4250	5/19/1998	male
00448	00449	4033690D44	*	3700	5/19/1998	male
00343	00726	4033730370	617	4100	5/20/1998	male

*not recorded

Table 1.2. Spawning matrix for 1998 indicating tag numbers for males and females, dates of mating, number of eggs produced and hatcheries where eggs and fry were incubated.

Date	**Female**	**Male**	**# Eggs**	**Hatchery**
5/21/1998	409/410	142/560	1450	McD=750, WS=700
5/22/1998	409/410	66/450	44040	UGA=17640, McD=9300, WS=10500, DC=6600
5/19/1998	585/600	448/449	9615	WS
5/20/1998	585/600	92/93	5327	WS
5/21/1998	563/591	559/587	3250	McD=2000, WS=1250
5/22/1998	563/591	570/571	23055	Mcd=7685, WS=8265, DC=7105
5/22/1998	563/591	539/561	30690	McD=10230, WS=10075, DC=10385
5/20/1998	123/730	343/726	3500	McD=1250, WS=2250
5/21/1998	123/730	301/586	9135	McD=4640, WS=4495
5/22/1998	123/730	489/490	12600	McD=3300, WS=5100, DC=4200
	TOTAL		**142,662**	

McD = McDuffie State Fish Hatchery
WS = Warm Springs National Fish Hatchery
UGA = University of Georgia, D.B. Warnell School of Forest Resources, Whitehall Fisheries Lab
DC = Dennis Wildlife Center, Bonneau, South Carolina

Incubation

Total egg production for 1998 was 142,662, with 17,640 being shipped directly to the University of Georgia for research projects, and 125,022 designated for the fingerling production program (shipped to Warm Springs National Fish Hatchery, Dennis Wildlife Center, Bonneau, South Carolina and McDuffie State Fish Hatchery). Embryos were transferred to McDonald hatching jars for incubation. Once hatching began, yolk-sac fry were transferred to aquaria. When the incubating fry reached swim-up, they were provided with freshly hatched *artemia* nauplii. When all fry began feeding consistently, they were transferred to one of seven State hatcheries in Georgia and South Carolina for pond rearing to Phase I fingerling stage. A total of 55,683 fry were shipped, with Warm Springs National Fish Hatchery retaining 2,000 for intensive rearing studies. Table 1.3 summarizes the 1998 spawning effort and compares results to previous years, including the numbers of fingerlings ultimately produced from spawned robust redhorse.

Table 1.3. Summary of the number of robust redhorse broodfish spawned, eggs, fry and fingerlings produced from spawning at the temporary hatchery facility adjacent to the boat ramp at the Beaver Dam Wildlife Management Area, Oconee River, Georgia during spring 1995, 1996, 1997 and 1998.

Year	Males spawned	Females spawned	Eggs produced	Fry produced	Fingerlings produced
1995	20	20	652,750	71,769	40,468
1996	21	11	477,119	98,089	1
1997	21	12	360,219	189,167	36,285
1998	10	4	142,662	55,683	13,030

Task 2. Effects of Temperature and Water Flow on the Incubation and Survival of Robust Redhorse *(Moxostoma robustum)* Eggs and Larvae-1997 (Year Three)

Since the discovery of the Oconee River population of robust redhorse Moxostoma robustum in 1991, spawning activity has been observed in late spring of each year at sites between Milledgeville and Dublin, GA. Fertilized robust redhorse embryos have been collected from the gravel substrate at spawning sites, but ichthyoplankton sampling subsequent to spawning observations has produced low and variable numbers of larval robust redhorse. Whether the sampling methods used to collect larvae provide an accurate estimate of larval abundance in the river is unclear; however, if these low estimates of larval density are representative, they may account for the paucity of juvenile robust redhorse collected despite considerable sampling effort in the main channel of the river. The successful production of healthy robust redhorse juveniles under carefully-controlled laboratory conditions suggests that environmental conditions may not be suitable for incubation of embryos and larvae to the point of emergence from the substrate.

This study was designed to establish optimal conditions of temperature and water flow for the incubation of robust redhorse embryos and larvae. Specific objectives of this study were to: 1) examine incubation success at a range of flow rates (turbulence levels), 2) examine the effects of water temperature (within the range likely to be encountered) on incubation success, and 3) evaluate possible effects of interactions between temperatures and flow rates on incubating embryos and larvae. This study was originally proposed in 1998 based on results of work funded by Georgia Power Company and conducted in 1996 and 1997. However, severe flooding during spring of 1998 made obtaining the embryos needed for this study impossible. Accordingly, work scheduled for this project will be rescheduled for the spring of 1999.

Task 3. Reproductive and Recruitment Success of Robust Redhorse in the Oconee River, Georgia.

Introduction

The arrival of the 1998 robust redhorse spawning season was met with much anticipation because of the sudden increase in the number of larval robust redhorse captured during the 1997 spawning season. Further, the first-ever capture of a larval robust redhorse in a light trap was considered additional evidence of a discernible increase in the abundance of larval robust. The dramatic increase in the abundance of larval robust redhorse was attributed, in part, to a new run-of-river flow regimen implemented during the 1997 spawning season (Jennings et al. 2003).

Thus, the approach of the 1998 spawning season allowed researchers evaluating the status of robust redhorse in the Oconee River to see if run-of-river flows during the spawning season would produce results similar to those observed during 1997.

Larval abundance data for 1998 were envisioned to provide evidence with which to evaluate hypotheses advanced to explain the apparent scarcity of larval robust redhorse and a population comprised of mostly large, old individuals (Evans 1994). If larval robust redhorse were at least as abundant during 1998 as they were during 1997, then unstable flows during previous spawning seasons might be advanced as a hypothesis explaining the apparent scarcity of larvae and juvenile robust redhorse. Otherwise, other hypotheses such as unsuitable spawning or rearing habitat, ontogenetic shifts in habitat use, or predation by flathead catfish remained viable explanations for the unusual demographics of the robust redhorse population in the Oconee River.

As in previous years (1995-1997), the goals of this work are to evaluate the reproductive and recruitment success of robust redhorse in the Oconee River. Specific objectives include documenting the abundance (i.e., density) and distribution of larval robust redhorse in the Oconee River from Milledgeville to Dublin, GA.

Material and Methods

Ichthyoplankton sampling

Larval and juvenile fishes were sampled from about a 51-km reach of the Oconee River that runs from Avant's Landing (N 32°56.35' W 83°03.97'), which is adjacent to the Thiele Kaolin mine near the Baldwin-Washington County line, downstream to the boat ramp at Beaverdam Wildlife Management Area (N 32° 40.54' W 82° 56.03') in Wilkinson-Laurens Counties. Sampling was begun on May 11 and continued to September 23,1998. Pushnets, light traps, and seines were used to sample larval fishes from the study reach; hoop nets were added to the gear repertoire to enhance the probability of capturing juvenile robust redhorse. The frequency and duration of sampling as well as habitats sampled with each gear varied (Table 3.1) in response to expected ontogenetic shifts in habitats by larval robust redhorse. Pushnets (505 μm) sampled ichthyoplankton drift about 1 m below the surface in the mainstem areas of the river, and light traps were used to sample ichthyoplankton from slack-water habitats. A seine (800 μm) was used to sample larval and juvenile fishes from sandbar and oxbow areas. Hoop nets (0.61- and 0.91-diameter) were used to sample deepwater habitats in the mainstem of the Oconee River, especially in deep meander sections that could not be sampled with seines.

Table 3.1. Gear types and deployment information for sampling larval and juvenile robust redhorse in the Oconee River, Georgia between May and August 1998.

Gear (mesh μm)	Dates	Effort	Frequency	No. of samples	Habitat sampled
push-nets[1] (505)	5/11/98 - 7/29/98	6 tows	2 x's /week	63	mid-channel, deep pools (occasionally)
light traps[2] (N/A)	5/25/98 - 7/29/98	6-10 traps	2 x's /week	108	slack water habitats off main channel, mouths of creeks (occasionally)
seine nets[3] (800)	5/29/98 - 9/23/98	9 hauls	1 x /week May - July, September; 3x's /week August	174	sandbars, mud-flats (occasionally)
Hoop-nets[4]	7/25/98 - 9/2/98	6 nets	2 x's /week	54	deep pools by woody debris

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[1] Mean water volume sampled was 100 m^3

[2] Light traps were fished for an average of 3-5 hours

[3] Seine hauls were on average 20 m long with a distance from shore of 5 - 10 m

[4] Two net sizes. Three 0.61 m-diameter nets, and three 0.91-m diameter nets

Ichthyoplankton samples were preserved in 10% formalin and transported to the University of Georgia where fishes and eggs were removed and enumerated; subsequently, all fishes were identified to the lowest taxonomic level possible. Twenty percent of the sorted samples were resorted to assess the efficiency of the extraction of larval fishes and eggs; this quality assurance sorting was conducted by someone other than the person who sorted the sample originally.

Water quality measurements

Environmental conditions (e.g., depth, flow, turbidity, water temperature, and dissolved oxygen) were measured at the time of sampling at each station. Temperature and dissolved oxygen were measured with a YSI ®Model 57 dissolved oxygen/temperature meter, turbidity was measured with a Hach ® Model 2100P Turbidimeter, flow was measured with General Oceanics ® flow meter, and depth was determined with a Lowrance ® boat-mounted depth finder.

Results and Discussion

Three hundred ninety eight samples containing 23,769 fishes from at least 12 families were collected during the 1998 field season (Table 3.2). Estimated efficiency with which fish and eggs were extracted from the samples was greater than 99%. Once again, seines seemed to be the most effective method for sampling larval fishes in the Oconee River. Light traps and push nets were about equally effective, but were much less effective than seines.

Generally, the number of families caught during1998 (12+) was similar to previous years, as was their relative contribution to the total catch (Table 3.3). Minnows (Cyprinidae) continued to dominate (91%) the catch, and most of the other families each contributed <1% (Table 3.3). The notable exception to this pattern was during 1995, when suckers (Catostomidae) and sunfish (Centrarchidae) each comprised > 5% of the catch and shad (Clupeidae) comprised > 2% of the catch. These differences in catch may be related to inter-annual variability in year class strength.

Robust redhorse spawned successfully during 1998, but the larval density (Figure 3.1) and number (Table 3.4) were lower than were measured during 1997. A total of six larval robust redhorse were captured during 1998; all were captured in pushnets (Table 3.4). Four of the six were captured in two different samples at the Avant's Landing spawning site on May 11, 1998. The fifth was caught on May 15th at Station 3, which is just upstream of Commissioner Creek, and the sixth was caught on May 25th in Station 2, in the area referred to as the "primitive boat ramp" (Figure 3.2). Larval robust redhorse were most abundant at the Avant's Landing site (maximum density = 10.1 larvae per 1000 m^3). Though lower than at the maximum density recorded at the Avant's Landing site, abundances of larval robust redhorse at Station 3 (3.3 larvae per 1000 m^3) and Station 2 (3.1 larvae per 1000 m^3) were comparable (Table 3.5).

The diversity and abundance of other larval and juvenile suckers during 1998 seemed to be comparable to levels documented in previous years (Table 3.4). Generally, carpsuckers *Carpoides* spp. dominated the catch, and spotted suckers *Minytrema melanops* were the second most abundant of the various suckers in the catch. Notch-lip redhorse

Table 3.3. Number of larval and post-larval fishes, by family (and percent catch composition), collected from the study reach of the Oconee River, Georgia during late spring and summers, 1995 - 1998.

	Number of fishes per family (and % catch composition)														
Year	Lepisosteids	Clupeids	Cyprinids	Catostomids	Ictalurids	Esocids	Belonids	Aphredoderids	Atherinids	Percichthyids	Centrarchids	Percids	Poeciliids	Soleids	Unknown
	4	1,041	35,658	3,468	243	0	16	0	739	0	2,675	94	956	1	282
1995	(<1.0)	(2.3)	(78.0)	(7.6)	(<1.0)	(0.0)	(<1.0)	(0.0)	(1.6)	(0.0)	(5.8)	(<1.0)	(2.1)	(<1.0)	(<1.0)
	3	393	35,373	705	92	0	6	0	311	0	459	151	688	2	532
1996	(<1.0)	(1.0)	(91.4)	(1.8)	(<1.0)	(0.0)	(<1.0)	(0.0)	(<1.0)	(0.0)	(1.2)	(<1.0)	(1.8)	(<1.0)	(1.4)
	2	938	19,128	261	38	1	1	0	187	0	169	143	680	1	46
1997	(<1.0)	(4.3)	(88.6)	(1.2)	(<1.0)	(<1.0)	(<1.0)	(0.0)	(<1.0)	(0.0)	(<1.0)	(<1.0)	(3.1)	(<1.0)	(<1.0)
	3	323	21,421	999	211	0	4	3	45	8	109	107	186	0	236
1998	(<1.0)	(<1.0)	(91.0)	(<1.0)	(<1.0)	(0.0)	(<1.0)	(<1.0)	(<1.0)	(<1.0)	(<1.0)	(<1.0)	(<1.0)	(0.0)	(<1.0)

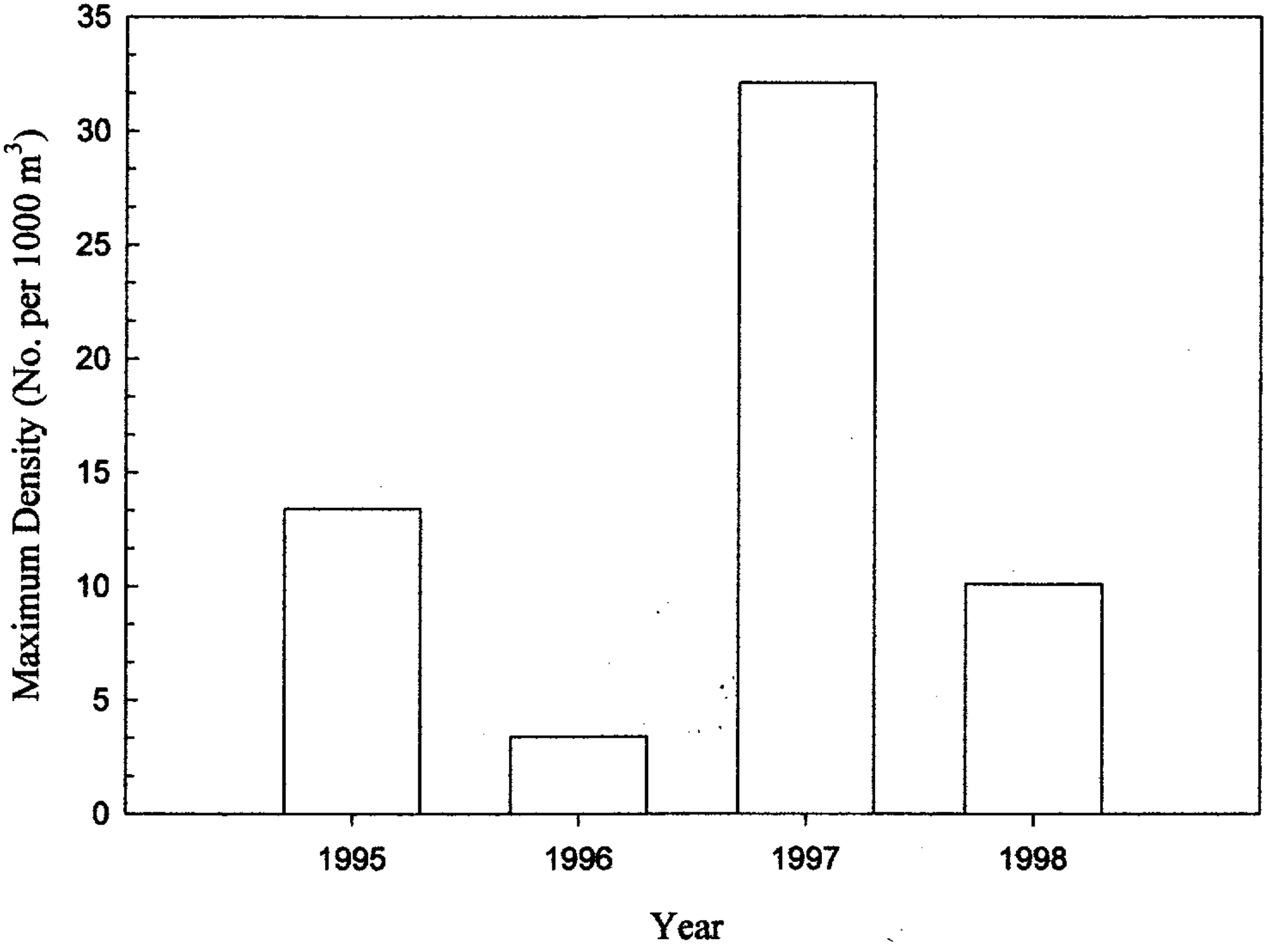

Figure 3.1. Maximum densities of larval robust redhorse collected from the Oconee River between Avants' Landing and Dead River during spring 1995-1998.

Table 3.4. Number of larval and post-larval Catostomidae collected from the Oconee River, GA during spring and summer 1995 - 1998.

	Species						
Year & gear	robust redhorse	silver redhorse	spotted sucker	carpsucker[5]	creek chubsucker	northern hogsucker	unknown
1998							
push - net	6	0	8	3	0	1	0
light trap	0	0	0	0	0	0	0
seine	0	2	38	941	0	0	0
1997							
push - net	19	5	13	32	1	0	0
light trap	1	0	29	1	0	0	1
seine	5	5	3	146	0	0	0
1996							
push - net	3	0	0	103	0	0	1
light trap	0	0	0	9	0	0	2
seine	0	36	0	546	1	0	0
benthic	4	0	0	0	0	0	0
1995							
push - net	5	0	2	243	0	0	26
light trap	0	0	1	90	0	0	2
seine	0	5	0	3,019	0	0	0
D - ring	1	0	3	65	0	0	6

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[5] Two undescribed species of carpsucker *Carpoides* occur in the sample reach of the Oconee River. One is related to the quillback carpsucker *C. cyprinus* and the other is related to the highfin carpsucker *C. velifer*.

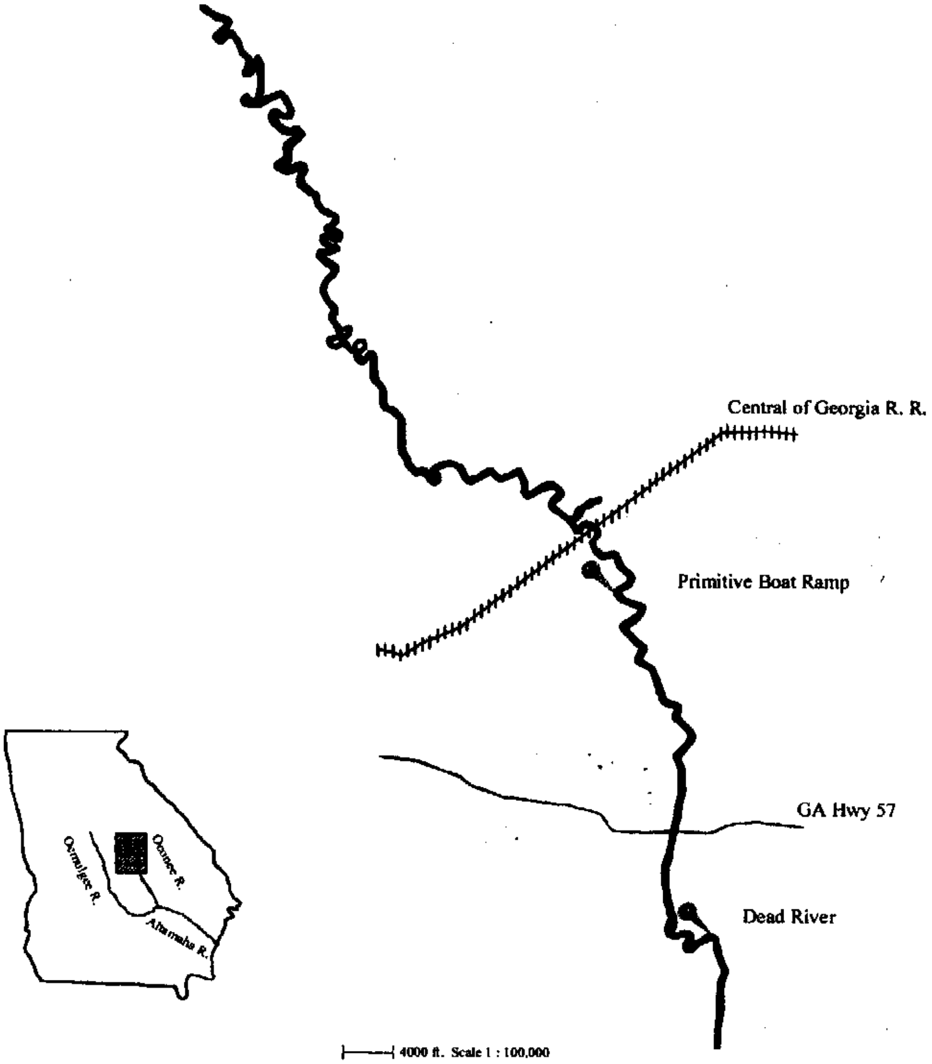

Figure 3.2. Map showing location of the "primitive boat ramp" and "Dead River" suspected spawning sites for robust redhorse in the Oconee River.

Table 3.5. Average total length (mm) and estimated density of larval robust redhorse *Moxostoma robustum* at specific reaches in the Oconee River between Milledgeville and Dublin, GA. 1998.

Date (1998)	Location (station)	Gear type	Time	Water temp (°C)	No./mean length (mm)	Estimated Density (larvae/1000 m^3)
May 11	Avants	Push-net	1153	20.6	1/13.7	3.4
May 11	Avants	Push-net	1203	20.6	3/14.5	10.1
May 15	3	Push-net	1310	22.0	1/12.4	3.3
May 25	2	Push-net	2050	26.1	1/14	3.1

Moxostoma collapsum, know previously as silver redhorse (R. Jenkins, Roanoke College - personal communication), were a bit less abundant (i.e., only two captured) during 1998 than in previous years (range: 5-36; Table 3.4, but look for silver redhorse in Jennings et al. 1996, 1998; 2003).

The abundance and distribution of juvenile robust redhorse in the Oconee River remain a enigma, as none were captured during 1998. Further, this absence from the sampling gear included the catch of hoop nets used to sample juvenile robust redhorse from areas not sampled effectively with the other gear used in this study. The hoop nets contained a total of 58 flathead catfish *Pylodictus olivaris*, none of whose stomachs contained juvenile robust redhorse or any remains that might be a diagnostic taxonomic clue (i.e., intact carcass or molariform gill arches) for robust redhorse. In addition to flathead catfish, the hoop nets also contained channel catfish *Ictalurus punctatus* (n=31), various sunfishes *Lepomis* spp. (n=22), common carp *Cyprinus carpio* (n=1), longnose gar *Lepisosteus osseus* (n=1), and one fish whose remains were unidentifiable.

The scarcity of larval robust redhorse and the limited distribution during 1998 compared with that observed during 1997 suggest that robust redhorse experienced lowered reproductive success during 1998. This scarcity was disappointing given the apparent success of the previous spawning season when larval robust redhorse were more abundant and distributed over a wider spatial area than in previous years (Jennings et al. 2003). Much of the credit for the increases in larval robust redhorse abundance was attributed to the run-of-river flows during the spawning season. Flows during the 1998 spawning season also were run-of-river, but the spawning results were not similar to the previous season.

Run-of-river flows may be one of a suite of factors that affect reproductive success of robust redhorse. Flows during May 1997 and May 1998 fluctuated less than flows during May 1996 and 1995. In fact, flows during May 1998 actually fluctuated slightly less than flows during May 1997 (Figure 3.3). Magnitude of flow was the primary difference between 1998 and 1997 run-of-river flows; the river was much higher for a longer duration during May 1997. Interestingly, larval abundance and distribution seem to benefit from the relatively stable low flows (Figure 3.3; top) that occurred during May 1997; however, the relatively stable high flows (Figure 3.3; bottom) that occurred during May 1998 did not produce similar results.

The cause(s) for the poor catches of robust redhorse and notch-lip redhorse during 1998 is unknown. The Oconee River experienced bank-full flows during much of the spring, and sampling was difficult. The swift currents during the flooding rendered light traps impractical. Also, the high water reduced the availability of sandbars to sample with seines. Despite the difficult sampling conditions, other suckers were sampled during 1998. Spotted suckers were at least as abundant as they were during 1997 and 1996, and carpsuckers were very abundant (>900 caught; second highest catch since 1995). Deploying and retrieving the hoop nets did not seem to be similarly affected by the high, fast water, but whether those conditions affected the behavior of fish is unknown. At least six species were caught in the hoop nets, but suckers were not among the catch. Apparently, all suckers do not benefit similarly from the same environmental conditions.

Redhorses generally spawn in shallow (< 1.5 m) water over gravel bars (Jenkins and Burkhead 1994), but whether depth alone is a factor in their spawning is unknown. In the Oconee River, the one confirmed (Avant's Landing site) and two suspected (primitive boat ramp

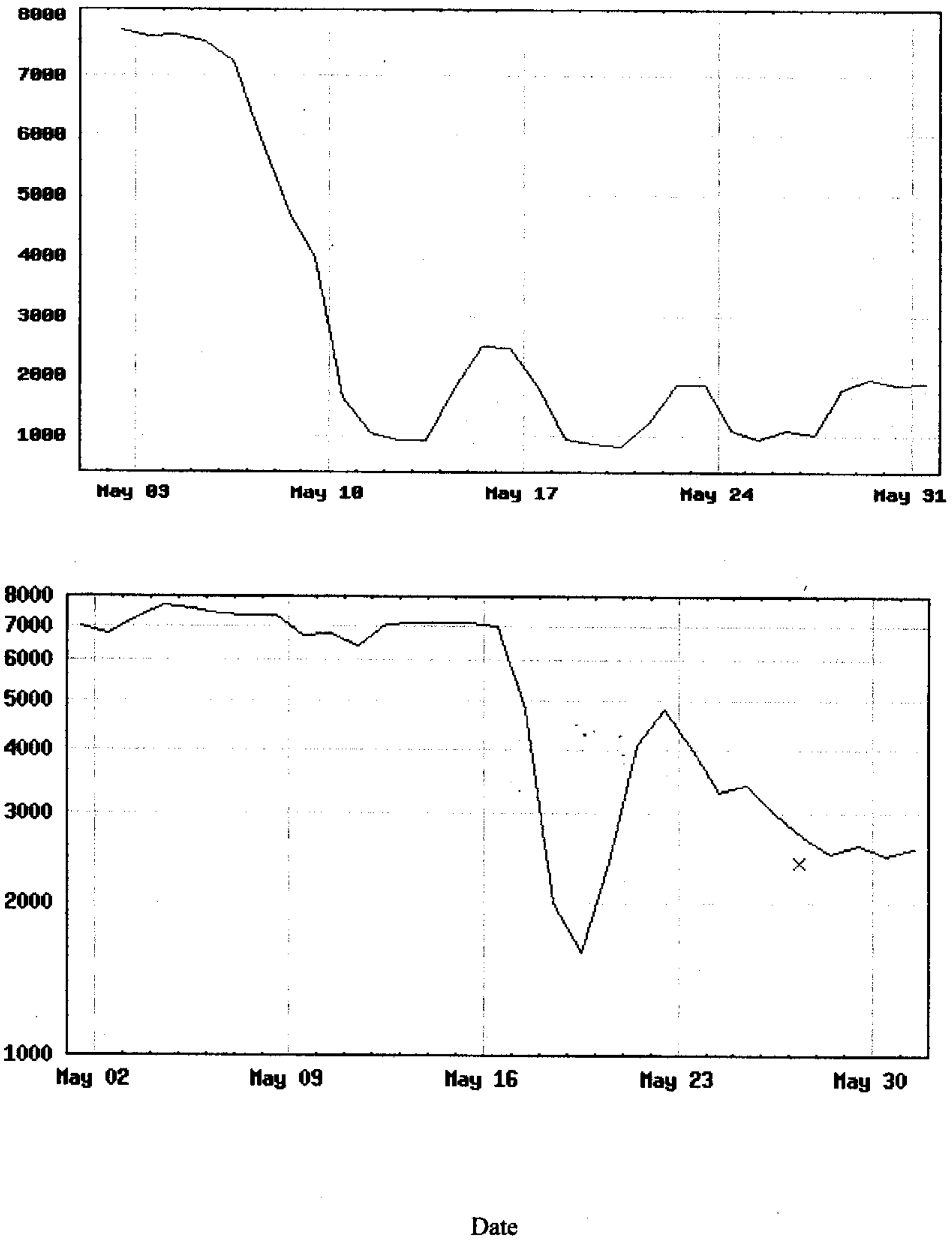

Figure 3.3. Mean daily discharge (x=measured flow; red = estimated flow) in the Oconee River during May 1997 (top) and 1998 (bottom) at the Avant Mine site where robust redhorse spawn (data provide by US Geological Survey, Gauge No. 0223056).

and dead river sites) spawning sites generally fit the "shallow water over gravel bars" description. The collection of a total of six larval robust redhorse from two sites in the Oconee during the high flows that occurred in the Oconee during May 1998 indicates that some spawning occurs in deeper water. However, the degree to which spawning behavior may be affected by the increased current velocity that accompanies high flow conditions is unknown. Metabolic costs associated with maintaining position over the gravel substrates would probably increase with flow. Whether there is an "abandonment threshold" above which the fish abort their spawning attempts is unknown, but similar environmental cues have been reported for species such as striped bass *Morone saxatilis* (Hill et al. 1989), paddlefish *Polyodon spathula* (Russell 1986), and lake sturgeon *Acipenser fulvescens* (Kempinger 1988). In addition to possible abandonment thresholds for adults, there probably are current velocities above which the spawning robust redhorse would encounter difficulties depositing fertilized eggs in gravel substrates.

Whether either of these two scenarios contributed to the apparent scarcity of robust redhorse during the high, relatively stable run-of-river flows that occurred during 1998 is unknown, but the pattern of larval abundance might offer a clue. The maximum density of larval robust redhorse occurred almost two weeks earlier during 1998 (May 15) than in previous years (~May 27). Early emergence of larval robust redhorse in a laboratory setting has been attributed to excessive fine sediment infestation (see Task 4 - this volume). This factor did not seem to be at work during 1998 because the larvae in the "early samples" were in the size range (i.e, 12-14 mm TL) of robust redhorse normally seen in push net samples. The size range of these larvae suggests they did not emerge early from gravel substrates. Instead, robust redhorse may have spawned successfully during early May 1998, but as the high flows persisted, the spawning fish may have abandoned the attempt or were unsuccessful at depositing fertilized eggs in the gravel.

Conclusions

Six larval robust redhorse were collected during the spring 1998, which was the second year of run-of-river flows during the spawning season. However, fewer larvae taken from a geographically smaller area compared with the previous year suggest that low stable flow produce promotes better reproductive success. For example, the catch of larval robust redhorse during 1998 when flows were high and fairly stable was comparable to catches during 1995 and 1996 when power generation and the resultant highly variable flow occurred during the spawning season. The mechanism responsible for the diminished reproductive success, as evidenced by low larval abundance is unknown. There may be a current velocity threshold above which the metabolic costs of residing over the spawning substrates are too high and the fish abandon attempts to spawn. A similar current velocity threshold may exit, and it may prohibit successful deposition of fertilized eggs into the gravel. Regardless of the mechanism, the available evidence, though limited, seems clear: robust redhorse seem to experience better reproductive success when flows are stable and relatively low (i.e., 2500 cubic feet per second).

Task 4. Effects of gravel quality and percent fine sediment on the hatching success of robust redhorse eggs

Introduction

Robust redhorse in the Oconee River spawn between late-April and early-June when water temperatures are between 21 - 25 °C (Looney 1997). Robust redhorse spawn in triads, composed of one female flanked by two males, over a loose gravel substrate in moderate to swift current (described in Jennings et al. 1996). While releasing gametes, the spawning trio quiver energetically and plow the substrate with their caudal and anal fins. This process results in egg burial and is consistent with observations of other species in the genus *Moxostoma* (Burr and Morris 1977; Jenkins and Jenkins 1980; Kwak and Skelly 1992). Larval robust redhorse hatch and remain in the interstitial spaces of the gravel substrate until most of the yolk sac has been absorbed. Upon reaching about 13 mm TL, the larvae emerge from the gravel to begin exogenous feeding (Jennings et al. 1996).

Robust redhorse spawning has been observed at one site in the Oconee River (Avant Mine boat ramp, Washington Co.), and evidence suggesting that spawning has occurred at another site (river mile 103, downstream of Avant) has been found (Jennings et al. 1996) (Figure 3.2). Other areas of gravel substrate similar in composition to that at Avant are also known to occur within the range of the Oconee population (EA 1994a and 1994b). Robust redhorse eggs have been collected from the gravel substrate at the Avant site, suggesting that spawning adults are successful in depositing fertilized eggs in the gravel (Jennings et al. 1996). However, estimates of larval abundance remain low (Jennings et al. 1998). Whether the absence of larvae in samples is related to their actual abundance or sampling inefficiency (e.g., gear or habitat) is unknown. As a result, questions exist about survival to emergence (STE) of larvae from the gravel substrate in which the eggs are deposited.

The present condition of many streams in the Midwest and Southeast is drastically different from that which existed prior to European settlement and agricultural development (Fajen and Layzer 1993). Such is the condition of the Oconee River. Since the turn of the 19th century, poor silvicultural and agricultural practices throughout the Oconee River watershed have drastically increased the sediment load of a river (Luft 1986, EA 1994a) that presumably, like other Piedmont/Upper Coastal Plain rivers, once ran "clear as crystal, their bottoms covered with a coarse gravel" (Byrd 1728 *in* Rowalt 1937). The Oconee River was still recovering from this massive sediment increase, which peaked in the early 1900s (Luft 1986), when Sinclair Dam was constructed in 1958. The reservoir formed by Sinclair Dam acts as an efficient sediment trap, and the river seems to have equilibrated with its present sediment load, transporting an amount of sediment equal to that entering the river (EA 1994a). However, the current bed load of the Oconee River is probably still representative of historical inputs of excessive fine sediment, and the gravel deposits used by spawning robust redhorse are typically composed of 25-50% fine sediment (EA 1994a and 1994b).

The effects of sediment on reproductive success of coldwater fishes is well documented, but such literature is scarce for warmwater fishes such robust redhorse (Muncy et al. 1979, Waters 1995). The available literature on how sediment contamination affects reproductive success of cold- and warm-water lithophilic spawners is reviewed in Jennings et al.(2003). Generally, sediment contamination is linked to several characteristics of the gravel beds in which these fishes spawn: 1) the availability and size of interstitial voids for developing eggs and larvae to reside within, 2) sufficient gravel permeability, such that hyporheic flow may supply developing eggs and larvae with dissolved oxygen and disperse metabolic wastes, and 3) a lack of consolidated sediments, which could impede or prevent larval emergence. These characteristics are directly influenced by the availability of suitably sized gravel particles (i.e., gravel quality) and the amount of fine sediment (i.e., clay, silt, and sand particles) present.

STE of larval robust redhorse is dramatically reduced at fine sediment infestation levels of 25 % or higher (Jennings et al. 2003). However, that threshold at which the STE is affected is unknown. The present study expands on this earlier research and attempts to better determine the threshold at which fine sediment infestation (i.e., between 5 - 25%) in gravel substrates adversely affect the STE of larval robust redhorse.

METHODS

Gravel Treatments

Commercially available river rock was washed and shaken through a series of five standard sieves (2.0-, 8.0-, 16.0-, 25.0-, and 37.5-mm) to obtain four size classes of gravel. These separate classes of gravel were combined to create a control gravel mixture that reflected the relative abundances of the size classes of gravel found at known or suspected robust redhorse spawning sites in the Oconee River, as ascertained from existing data (EA 1994a and 1994b) and preliminary field surveys. This mixture consisted (by volume) of 25.0-37.5 mm (10%), 16.0-25.0 mm (40%), 8.0-16.0 mm (30%), and 2.0-8.0 mm (20%) diameter gravel. Sufficient amounts of commercially available concrete sand (particles < 2.0 mm in diameter), chosen as a good representation of the fine sediment found in the Oconee River, were added to the control gravel mixture to produce four experimental gravel mixtures that contained 0, 5, 10, 15, 20 and 25% fine sediment by volume. There were four replicates of each experimental mixture.

Experimental Apparatus

The following brief account of the experimental apparatus is presented here for convenience. A detailed account of the construction and dimensions of the experimental apparatus is given in Jennings et al. (2003). The experimental apparatus consisted of a series of incubation cells housed in a large fiberglass trough with recirculating water flow and was constructed at Whitehall Fisheries Laboratory, University of Georgia. Incubation cells (Figure 4.1) consisted of 15.5-L opaque, plastic container. A polyvinyl chloride (PVC) standpipe placed in each of the cells allowed dissolved oxygen concentrations of water within the gravel matrix at the depth of egg burial to be monitored and was capped between observations to minimize surface–water gas exchange. Gravel mixtures were added to each cell until the depth was level with the perforations in the standpipe. The larger diameter pipe facilitated introduction of fertilized robust redhorse eggs. Portions of experimental gravel mixtures corresponding to the contents of each cell were retained in individual 1-L containers. Following egg introduction, this retained material was used to cover the eggs, and the large diameter standpipe was removed. A grid-like pattern of incubation cells and non-incubation cells, which did not contain perforations or eggs, was used to minimize channeling of water flow around the incubation cells (Figure 4.2). Gravel treatments were randomly assigned to an incubation cell within this pattern.

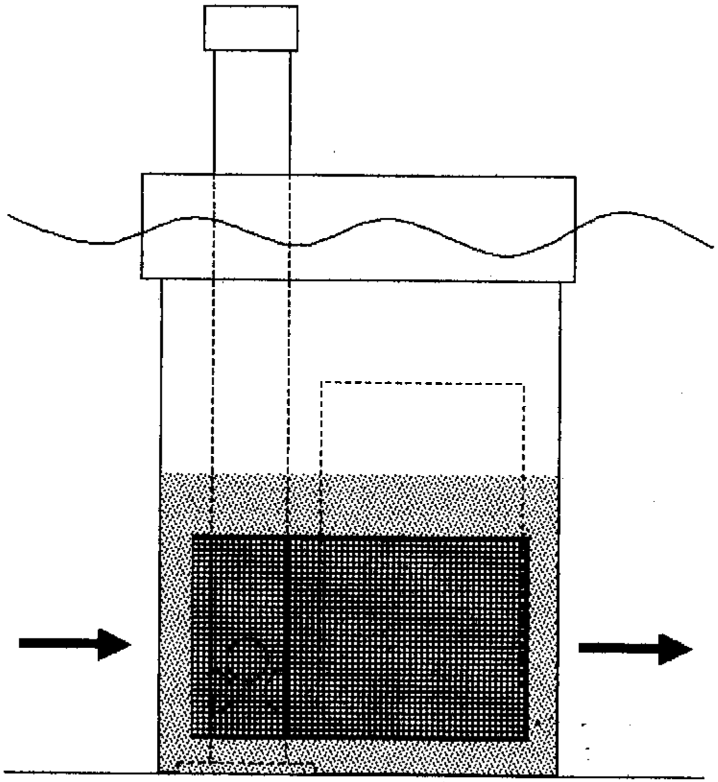

Figure 4.1. Diagram of incubation cell used to house gravel treatments and incubating robust redhorse eggs and larvae. The smaller diameter standpipe allowed dissolved oxygen concentrations to be monitored, and the larger diameter pipe facilitated egg introduction at a standard burial depth. Arrows denote direction of water flow.

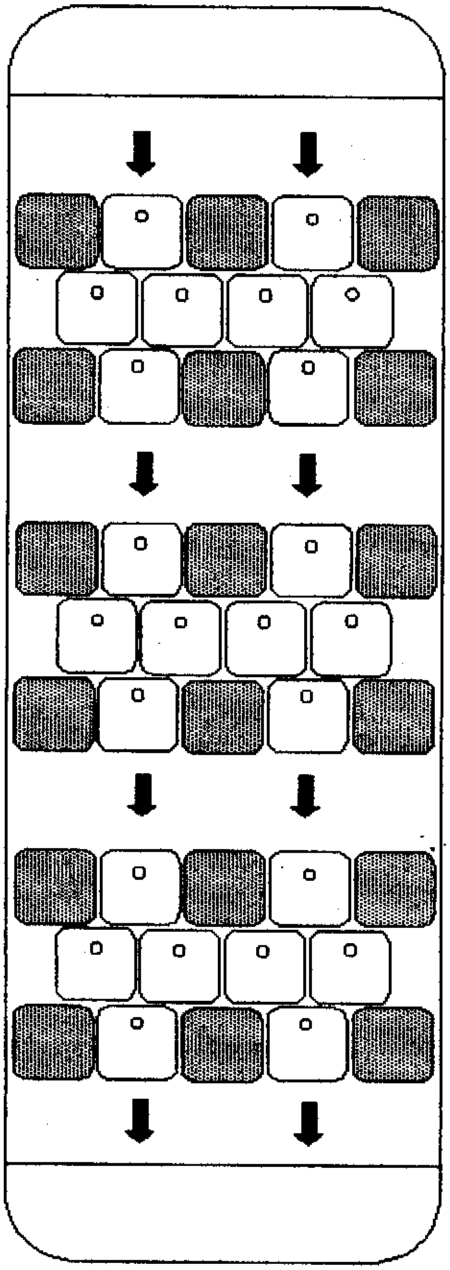

Figure 4.2. Grid-like arrangement of incubation cells (white) and non-incubation cells (shaded) used to test the effects of fine sediment and gravel quality on survival to emergence of larval robust redhorse. Circles represent standpipes used to monitor dissolved oxygen concentrations. Arrows denote direction of water flow.

Water flow, supplied by gravity return of water from the recirculation system head-tank, was supplemented with two submersible sump pumps in each trough. Acrylic baffles of differing heights created directional flow through the trough. Incubation cell height exceeded water depth and prohibited emergent larvae from leaving the cells. Water temperature was regulated between 21 and 23 °C with a 1-hp water chiller. These temperatures matched those commonly observed in the Oconee River during the spawning period of robust redhorse (Jennings et al. 1998).

Egg Introduction and Monitoring Protocol

Fertilized eggs were obtained from a single crossing of wild-caught brood stock that were spawned on May 22, 1998. The fertilized eggs were immediately transported to the Whitehall Fisheries Laboratory at the University of Georgia and placed in the incubation cells. The 1998 eggs were slightly larger (24.5 eggs/mL) than those used in the 1997 trials (27.9 eggs/mL). All fertilized eggs were buried at a depth of 5 cm. Because most of the larvae observed during the 1997 study emerged in the morning (Jennings et al. 2003), the experimental cells were observed once daily from 0800-1000 hours. Dissolved oxygen concentrations (mg/ L) within each cell were recorded with a calibrated YSI® Model 55 dissolved oxygen meter during each observation period.

Upon emergence from the gravel, larvae were collected and preserved in 10% formalin. Emergence was deemed complete when larvae did not emerge for two consecutive days following peak emergence. Preserved larvae were later enumerated and total lengths were measured to the nearest 0.1 mm with a dial caliper. Larval length at emergence was not evaluated in this year of study. All other experimental protocols were unchanged from the 1997 study (see Jennings et al. 2003).

Data Analysis

All data were tested for normality with the Shapiro-Wilkes test (SAS Institute 1990) and for homogeneity of variances with Hartley's F-max test (Sokal and Rohlf 1981). Results of the Shapiro-Wilkes test indicated that the larval STE data were not normally distributed. Transformation failed to normalize the distribution, and an F-max test indicated that the variances of the data set were homogeneous and the *F*-test is reasonably robust against non-normality (Netter et al. 1990). Therefore, mean dissolved oxygen concentrations among treatments were compared with ANOVA and least square means separation tests. A three-parameter sigmoidal regression model was used to describe the relationship between larval STE and percent fine sediment infestation during the 1998 study as well as the combined data for both study years (i.e., 1997 & 1998). An $\alpha = 0.05$ was used to evaluate the significance of all statistical tests.

RESULTS

Incubation Period and Timing of Larval Emergence

Larval emergence began 11 days after fertilization and peaked at days 16 and 17 post-fertilization. Low levels of intermittent larval emergence were observed until day 27 post-fertilization. Emerged larvae that emerged after day 11 swam actively throughout the water column enclosed by the incubation cells, but most aggregated near the surface. Larvae exhibited an avoidance response during collection periods, with some attempting to elude capture by seeking refuge in the substrate. Larvae either hid in the interstitial spaces of the upper substrate layer or hovered, motionless just above the substrate. The drab larval coloration served to effectively camouflage the latter against the substrate.

Larval Emergence

Mean larval emergence varied among the treatments and was highest (69.8%; S.E.=0.01) in the control mixture without fine sediment and lowest (9.1 %; S.E.=0.04) in the 25% fine sediment treatments (Figure x). Larval STE in the treatments containing 20 and 25% fine sediments was drastically lower than STE in the treatments contains <20 % fine sediments. An inverse relationship existed between percent fine sediment levels and larval STE ($P < 0.0001$). This relationship was non-linear ($P < 0.0001$) and was best described by a three-parameter sigmoidal model ($R^2 = 0.89$). Combining the Trial I emergence data from 1997 (see Jennings et al. 2003) and the current year resulted in a better fit of this model ($R^2 = 0.94$) (Figure 4.3).

The presence of 596 larvae outside the incubation cells indicate that lateral movement by pre-swim up larvae in the interstitial spaces occurred again during the 1998 study. The escaped larvae represented about 10% of the combined total number of eggs placed in all the trials. This loss was deemed tolerable, and escapement rate was assumed to be equally distributed among treatments and replicates.

Hyporheic Dissolved Oxygen Concentration

Mean dissolved oxygen concentrations in the treatments ranged from 7.8 (S.E. = 0.00) to 7.9 (S.E. = 0.029) and did not vary ($P = 0.1377$) over the course of the study (Table 4.1).

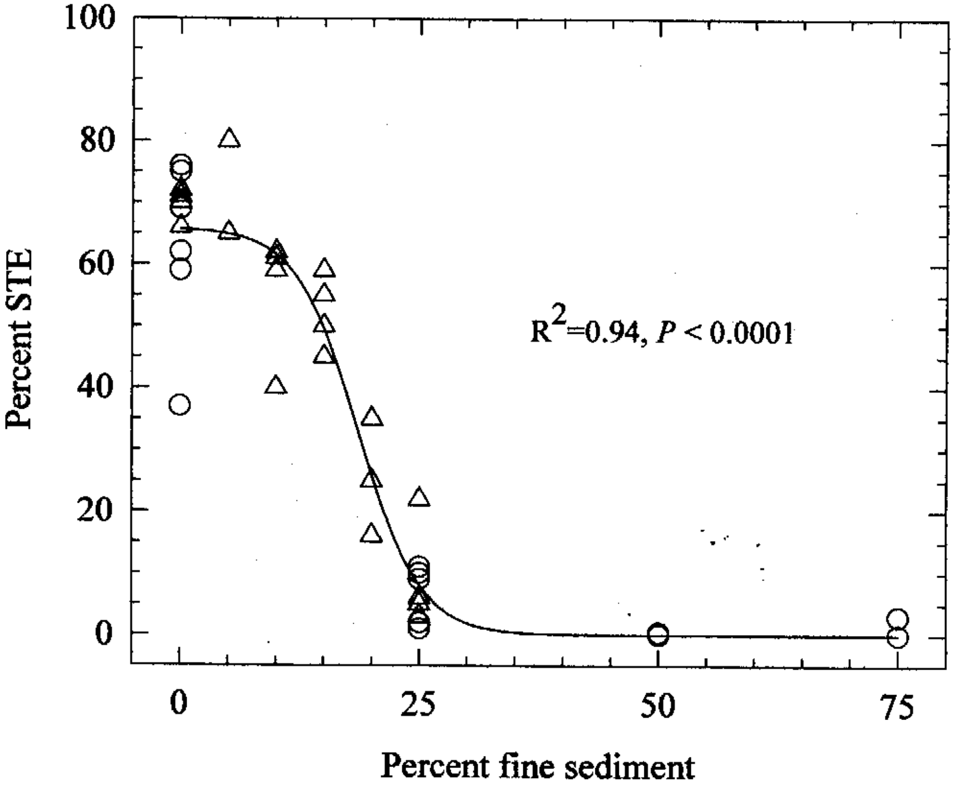

Figure 4.3. Effect of increasing levels of fine sediment (x) on larval robust redhorse survival to emergence(STE) rates (y). Emergence rates observed in individual replicates during 1997 are represented by (○) and those observed in individual replicates during 1998 are represented by (△).

Table 4.1. Mean dissolved oxygen (DO) concentration and associated standard error within each treatment that tested the effect of fine sediment on larval robust redhorse survival to emergence.

Fine sediment (%)	Mean DO (mg/L)	SE
0	7.9	0.029
5	7.8	0.000
10	7.8	0.025
15	7.9	0.029
20	7.8	0.025
25	7.8	0.025

Similarly, mean dissolved oxygen concentration did not differ ($P = 0.7006$) among the five treatment levels of fine sediment infestations (Table 4.1).

DISCUSSION

Effect of Fine Sediment on Larval Emergence

The inverse relationship between STE of larval robust redhorse and fine sediment pollution in rearing substrates reported previously (Jennings et al. 2003) was confirmed again this year, but with better resolution of the threshold at which STE is adversely affected. The nonlinear nature of this inverse relationship implies that robust redhorse larvae are reasonably tolerant of fine sediment until some threshold concentration (about 15% fine sediment) is reached. Above this threshold, larval emergence rates decline rapidly. Generally, the STE patterns observed for larval robust redhorse is consistent with that reported for salmonids (Phillips et al. 1975; Hausle and Coble 1976; Hall 1986; Chapman 1988).

The presence of yolk-sac larvae outside the incubation cells confirm that incubating larval robust do move laterally among interstitial spaces in gravel substrates. This phenomenon was observed during the first year of this study, but was attributed to the excessive amounts of fine sediments (Jennings et al. 2003). In the present study, fine sediment levels were much lower than those in the previous experiment. Early larval emergence of yolk-sac larvae from gravel substrates can have adverse effects on their survival, and the mechanisms for the lower survival of early-emerged are discussed in Jennings et al. (2003). Whether the lateral movement by the escaped larvae is simple preparatory behavioral in response to imminent ontogenetic shifts in habitat (i.e., preparing to swim up) or is in response to unfavorable microhabitat conditions associated with fine sediment pollution is unknown. If interstitial lateral movement is preparatory behavior for swimming up, then the small area of the incubation cell may explain why these larvae encountered the mesh screen and escaped. However, if the lateral movement is in response to excessive fine sediment, the threshold (i.e., 15%) at which reduced STE occurred in this study may provide a meaningful management objective to ensure continued or improved recruitment of larval robust redhorse in Oconee River.

Effect of Fine Sediment on Hyporheic Dissolved Oxygen Concentration

In the present study, dissolved oxygen concentrations remained well above the 5 mg/L recommended for survival and growth of most fishes (Piper et al. 1982). These results support conclusion from the 1997 study (Jennings et al. 2003), which state that ≥ 50% fine sediment infestation reduces dissolved oxygen concentrations compared to gravel substrates with ≤ 25% fine sediment load. The specific mechanism for this phenomenon is given in Jenning et al. 2003).

Incubation Period and Larval Behavior

Larval incubation and emergence periods in the present study were similar to those observed in the first year of this experiment (Jennings et al. 2003) and in other laboratory or hatchery conditions (Jennings et al. 1996). Generally, larval robust redhorse begin to swim-up about 11-14 days following fertilization, with peak emergence occurring about 2 - 4 days later. Generally, larval emergence is completed about 30 days post fertilization (Jennings et al. 2003). These data suggest that emergence of larvae in the wild may occur over a much longer time (i.e., on the scale of a month) than was suspected previously. Fluctuating river flows during and following the spawning period of robust redhorse might scour eggs and larvae from the substrate at a premature level of development (Montgomery et al. 1996) or result in further infiltration of fine sediments into the gravel matrix (Sear 1993). Therefore, our findings underscore the benefits of stable flows, adopted as part of the Federal Energy Regulatory Commission (FERC) re-licensing of Sinclair Dam, during the spawning and rearing season of robust redhorse in the Oconee River. Stable flows promote successful reproductive and recruitment success of robust redhorse in the system.

Newly emergent larval robust redhorse exhibited an escape response during collection periods in this study. The cryptic behavior of hiding in or near the substrate may serve some function in predator avoidance. Further, larval emergence seems to be a nocturnal phenomenon, which may be adaptive in that newly emerged larvae are given a chance to orient themselves and seek refuge in their lotic environment while sight-dependant predators are less effective.

Factors Affecting Larval Robust Redhorse STE

Increasing levels of percent fine sediment were inversely related to larval robust redhorse STE in the present study. Results for the first year of this study implicate low dissolved oxygen and entombment as the two primary mechanism responsible for the reduced larval STE (Jennings et al. 2003). In the current study, dissolved oxygen was well above the levels that affect fish survival; therefore, the declines in larval emergence observed in this study probably resulted from larval entrapment. These larvae probably developed normally, but were physically unable to emerge from the substrate.

Implications for Recruitment

This research implicates fine sediment pollution of gravel substrates as a significant threat faced by incubating robust redhorse eggs and larvae in the Oconee River. Spawning substrates used by robust redhorse in the Oconee River typically are composed of 25-50% fine sediment (EA 1994a and 1994b). At these fine sediment levels, emergence rates of less than 8.0% would be predicted by the model developed in this study. However, the percentage of fine sediment in spawning substrates is not uniform, and fine sediments are swept from the gravel substrates where robust redhorse deposit eggs (i.e., the sediment plume observed commonly during spawning). As a result, the extent to which robust redhorse recruitment in the Oconee River is constrained because of fine sediment infestation or whether the current level of larval recruitment is sufficient to maintain the population are unknown.

Runoff control and soil stabilization projects, particularly procedural controls (Wesche 1993), combined with effective interdiction measures have the potential to reduce the sediment load of the Oconee River and improve the spawning habitat available to adult robust redhorse. However, projects of this sort require time to reduce levels of deposited fine sediment. The uncertain status of the robust redhorse necessitates more immediate action. Restorative processes (i.e., the actual removal of excessive fine sediment from the spawning gravels) may be in order (see review by Waters 1995). Flushing flows and stream alterations can remove fine sediment from some areas, but may also increase fine sediment concentrations further downstream. Sediment traps and gravel cleaning devices, such as the "Gravel Gertie" (Mih and Bailey 1981) or hose and water jet (Waters 1995), may be a better solution as they can result in a net removal of fine sediment from the system. However, the application of restorative procedures to a large river such as the Oconee could prove difficult. Therefore, remediation procedures, such as constructing artificial spawning beds by gravel addition, should also be considered. The continued success of restoration and remediation measures relies on a decrease in fine sediment input; and therefore, the control of fine sediment addition through prevention and interdiction should be encouraged.

Reductions in STE associated with excessive amounts of fine sediment in the Oconee River could reduce larval robust redhorse cohorts to such small numbers that current production of juveniles would be extremely limited. Improvements in robust redhorse spawning habitat may increase the number of larvae that emerge successfully from the substrate, recruit to the population as juveniles, and improve the long-term prospects of a self-sustaining population of robust redhorse in the Oconee River and elsewhere.

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