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Hatchery rearing environment and age affect survival and movements of stocked juvenile lake sturgeon

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Abstract Considerable uncertainty exists over the relative merits of alternative supplementation strategies for lake sturgeon, *Acipenser fulvescens* Rafinesque. Numerous supplementation prescriptions have been advocated, largely in the absence of data, focusing on perceived impacts of levels of genetic diversity of progeny collected using different collection methods, and probabilities of survival relative to the size or age of fish released. The hypothesis that collection method, hatchery rearing environment and size/age at the time of release do not influence survival and movements was tested. *Acipenser fulvescens* gametes and juveniles were collected using three methods and reared in a stream-side hatchery along the natal stream and in a traditional non-natal hatchery environment. *Acipenser fulvescens* were released into the Upper Black River, Michigan at 8, 13, and 17 weeks of age. Higher rates of recapture were found for juvenile sturgeon reared in the stream-side hatchery than the traditional hatchery for the releases at 8 and 13 weeks. Recapture rates and dispersal distances were significantly greater for fish stocked at 17 weeks than for fish released at earlier ages. Large body size was negatively correlated with timing of movements across all ages indicating that survival may be enhanced by releasing individuals at night. Results indicate that supplementation protocols for *A. fulvescens* should be developed on a system specific basis and demonstrate the importance of hatchery-rearing environment when fish of younger ages are released.

KEYWORDS: *Acipenser fulvescens*, collection methods, dispersal, supplementation.

Introduction

Hatchery programmes have been widely used as both enhancement and management tools to address declines in wild fish populations (Waples 1999), but are receiving increased scrutiny because of the potential negative impacts of captive reared individuals on natural populations (Ford 2002; Miller *et al*. 2004; Araki *et al*. 2007). Criticisms have been raised based on empirical evaluations of the relative effects of different management options including; hatchery-rearing environment (Berejikian *et al*. 2000), age of release into the natural environment (Paragamian & Kingery 1992), and gamete, juvenile or broodstock collection and maintenance protocols (Flagg & Nash 1999).

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Stocking protocols developed from hatchery programmes are used to increase wild population abundance by increasing probabilities of survival during critical life history stages (Alverson 2002; Brown & Day 2002). Furthermore, hatchery programmes have the potential to supplement recruitment when unfavourable environmental conditions result in high embryo and larval mortality (Secor & Houde 1998). Although stocking programmes have been used to establish and enhance many fisheries, certain programmes have failed to increase the numerical abundance of the target species (Secor *et al.* 2000; Brown & Day 2002; Myers *et al.* 2004; Baer *et al.* 2007). General conclusions concerning the effectiveness of stocking programmes have not been reached, and challenges remain to reconcile benefits and potential costs to population dynamics, genetic integrity of resident populations, and to ecosystem processes and resource economics (Travis *et al.* 1998; Utter 1998). Supplementation programmes are becoming increasingly important for conservation efforts of threatened and endangered species (Brown & Day 2002), including regionally imperilled lake sturgeon, *Acipenser fulvescens* Rafinesque. Stocking prescriptions have typically been based on salmonid literature (Fraser 2008), although species such as *A. fulvescens* have very different life histories, mandating that research investigating the success of alternative methods of culture and release be conducted on this species explicitly.

Acipenser fulvescens is considered threatened throughout much of its range. This species has experienced dramatic declines in both numerical abundance and in distribution range. Its abundance in the Great Lakes has been projected to be < 1% of historical levels (Hay-Chmielewski & Whelan 1997). Current impediments to *A. fulvescens* recovery include the sensitivity of adults to anthropogenic factors such as over-harvesting, degradation in water quality and spawning habitat and loss of connectivity because of impoundment (Holey *et al.* 2000). In recent years, *A. fulvescens* recovery through stocking has become a high priority throughout the Great Lakes (Peterson *et al.* 2007). However, considerable uncertainty remains regarding the efficacy of different egg and larval collection methods, and the appropriate age of fish to stock. Success of supplementation programmes is typically based on estimates of contributions of stocked fish to older age classes (Li *et al.* 1996) or return to post stocking assessments (Walters *et al.* 1997). However, these methods are not amenable to *A. fulvescens* as they are not widely subject to harvest and younger age classes are not efficiently captured in surveys using standard sampling methods (Benson *et al.* 2005). Late age at maturity, infrequent spawning, low recruitment rates (Nilo *et al.* 1997), and occupancy of geographically separated breeding and non-breeding areas also confound interpretations, dictating that other methods be employed to evaluate supplementation programmes. In an extremely long-lived species such as *A. fulvescens*, survival during early-life stages represents a substantial component of variation in lifetime fitness, and therefore represents an opportunity for selection during these life stages. Evaluations of hatchery supplementation protocols would be most profitably focused on documenting factors related to survival immediately following release.

A number of factors contribute to the success of hatchery-based stocking programmes. Stocking of *A. fulvescens* throughout the Great Lakes region has employed numerous techniques for collecting progeny. Typically, gametes are removed directly from actively spawning adults (Folz *et al.* 1983; Ceskleba *et al.* 1985) or larvae are collected dispersing downstream from the spawning grounds (Auer & Baker 2002; Smith & King 2005). An alternative method, given the species broadcast spawning behaviour (Bruch & Binkowski 2002), was used by Crossman (2008) to collect eggs that were naturally produced and deposited on the stream substratum. Following collection and rearing, juveniles were stocked at different ages (Schram *et al.* 1999): larvae (13–25 mm) and juveniles (102–203 mm). Stocking studies that attempted to identify optimal ages for release into natural environments reported mixed results with respect to survival based on age at stocking (Elrod *et al.* 1988; Margenau 1992; Secor & Houde 1998; Amtstaetter & Willox 2004). Rearing hatchery fish to larger sizes leads to increased survival upon release (Gunn *et al.* 1987; McKeown *et al.* 1999; Yule *et al.* 2000), and probabilities of juvenile survival may be non-independent among members of the same family. Accordingly, if different gamete collection methods result in different levels of mean coancestry relative to the number of adults contributing to individuals that survive to the time of release, then evaluations of different gamete/larval collection methods are warranted. Recently, stream-side hatcheries have been advocated as a recovery tool for *A. fulvescens* throughout the Great Lakes (Holtgren *et al.* 2007). Stream-side hatcheries use water from streams targeted for release to maximise probabilities of imprinting and to help reduce the degree of domestication selection relative to traditional hatchery environments. Conservation hatcheries have been used for trout and salmon and have attributed positive results to more natural rearing conditions (Maynard *et al.* 1996; Berejikian *et al.* 2000). In the present study, the effects of different hatchery environments by rearing *A. fulvescens* are examined in two settings, a stream-side hatchery and a traditional hatchery, to quantify differences in post-stocking success.

The aim of this study was to assess the role *A. fulvescens* rearing conditions, collection methods, and size and age at stocking to determine probabilities of survival, and movements following release into the natural environment. The null hypothesis that gamete/larval collection method, hatchery rearing environment and age at release are not significant predictors of post-stocking survival for juvenile *A. fulvescens* was tested. Studies identifying the success of stocking strategies typically define success by the proportion of fish recaptured in subsequent assessments (Leber *et al.* 2005). This criterion was adopted and directed research objectives to: (1) determine contributions of fish stocked from two different rearing environments and three different collection methods on the rate of recapture using in-stream assessment; (2) determine which stocking age results in higher rates of capture (a surrogate measure of survival); and (3) quantify rates of downstream dispersal as a function of age and size.

Study site

Research was conducted on the Black Lake system (~3000 ha), Michigan (Fig. 1, see details in Smith & Baker 2005; Smith & King 2005). Fish were stocked into the Upper Black River, a fourth order stream located in the north-eastern corner of the state of Michigan that is highly influenced by dams. The hydrology of the Upper Black River provides an opportunity to enumerate a large proportion of the annual adult spawning run (Forsythe 2010) as well as to collect gametes and juveniles. Physical variables (water depth and flow; Table 1) provided wadeable conditions that permitted in-stream experimental work for several kilometres downstream of the first impoundment.

Materials and methods

Juvenile *A. fulvescens* were reared in two hatchery environments. Half of all fish were reared at a stream-side hatchery using water from the natal stream on the Upper Black River, Michigan. The remaining fish were reared using ground water at the Wolf Lake state hatchery (Michigan Department of Natural Resources) in southwest Michigan, which is a traditional hatchery environment. The stream-side hatchery used a flow-through design into which water was pumped directly from the Upper Black River (Fig. 1). Water was mechanically filtered to remove large sediments. Flow rates were kept constant and at a level that resulted in approximately two complete water turnovers every hour. Cleaning, feeding, lighting and water flow protocols in the two hatchery environments followed standard *A. fulvescens* rearing methods (Ceskleba *et al.* 1985; Fajfer *et al.* 1999). Fish in both hatcheries were kept in 1.22-m diameter tanks (0.5 m deep), which were divided to include two replicates of three different collection methods for various life stages. The first two methods, which are commonly used for many sturgeon species were direct egg takes (Birstein 1993) and collection of dispersing larvae (Auer & Baker 2002). The third method involved collecting eggs naturally fertilised and deposited on the stream substratum by spawning adults (Crossman 2008).

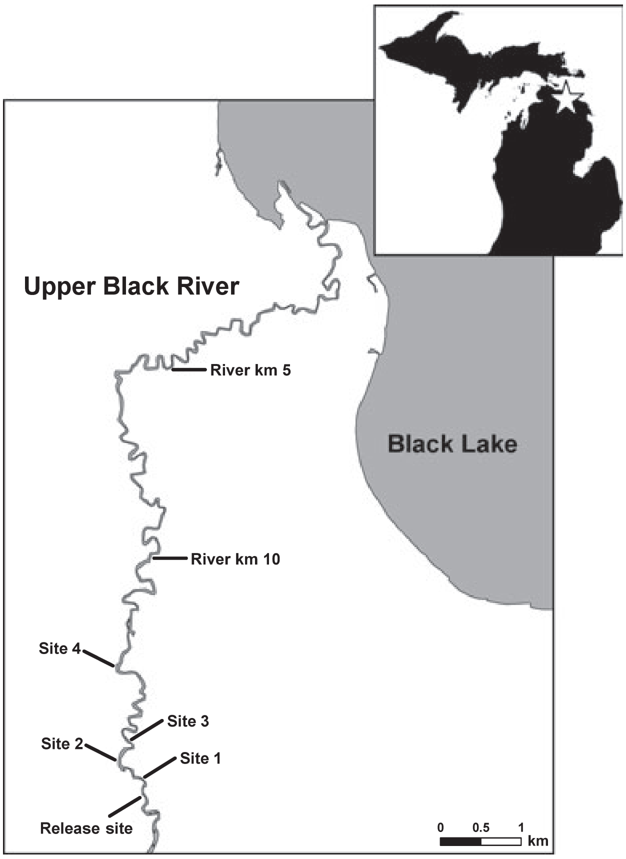

Figure 1. The Black Lake study site in Michigan, showing the release location and sampling sites (1–4) for release assessments in the Upper Black River.

Direct gamete takes (DGT)

Acipenser fulvescens were captured on the spawning grounds using large dip nets. Eggs were removed by

Table 1. Numbers released and total length (cm, mean ± 1 SE) corresponding to age, rearing environment, and gamete/juvenile collection method for three experimental releases of juvenile lake sturgeon into the Upper Black River, Michigan. Physical conditions including water depth (m) and velocity (m s^{-1}) are included for each downstream assessment site during each release. Letters (a, b, c) correspond to statistically greater proportions of fish recaptured between the different collection methods within each release age based on analyses of variance computed within a general linear mixed effects model

Age (weeks)	Hatchery environment	Collection method	Total released	Proportion recaptured	Total length, mm	Site	Water depth	Water velocity
8	Streamside	DGT*	1257	0.09^a	7.87 ± 1.06	1	0.62 ± 0.05	0.79 ± 0.04
		DL†	485	0.05^b	7.51 ± 0.82	2	0.52 ± 0.06	0.73 ± 0.03
		NPE‡	220	0.07^b	7.68 ± 1.11	3	0.61 ± 0.06	0.62 ± 0.04
	Traditional	DGT	1380	0.03^b	7.38 ± 0.99	4	0.74 ± 0.01	0.32 ± 0.02
		DL	725	0.04^b	7.42 ± 0.73			
		NPE	240	0.02^c	7.77 ± 1.22			
		Total	4307	0.05				
13	Streamside	DGT	999	0.16^a	10.84 ± 0.08	1	0.62 ± 0.13	0.74 ± 0.03
		DL	494	0.06^b	11.70 ± 0.20	2	0.68 ± 0.05	0.71 ± 0.10
		NPE	181	0.13^a	11.98 ± 0.17	3	0.77 ± 0.09	0.59 ± 0.03
	Traditional	DGT	623	0.10^a	11.20 ± 0.20	4	0.99 ± 0.07	0.40 ± 0.01
		DL	692	0.04^b	11.70 ± 0.14			
		NPE	237	0.10^a	12.90 ± 0.21			
		Total	3226	0.10				
17	Streamside	DGT	1000	0.16^a	13.85 ± 0.15	1	0.65 ± 0.06	0.48 ± 0.16
		DL	1088	0.08^b	14.81 ± 0.20	2	0.66 ± 0.03	0.45 ± 0.18
		NPE	399	0.26^c	14.49 ± 0.15	3	0.68 ± 0.03	0.46 ± 0.18
	Traditional	DGT	1000	0.17^a	16.51 ± 0.19	4	0.93 ± 0.05	0.37 ± 0.24
		DL	493	0.05^b	17.19 ± 0.30			
		NPE	208	0.27^c	15.80 ± 0.34			
		Total	4188	0.15				
		Grand Total	11721	0.10				

*Artificial: paternal half-sib families, each created with one female and two males (n = 26 & 25 for 2 years). †Drift: larvae captured dispersing downstream from spawning areas. ‡Natural: naturally fertilised and deposited eggs that were collected from the stream.

hand stripping females captured in the act of spawning. Eggs were placed in sealed plastic bags and transferred to a cooler to maintain ambient river temperatures. Milt was collected using a 30-mL syringe, then immediately placed on ice. Fertilisations were conducted within 12 h of egg collection. Egg volumes from each female were measured, divided into two equal lots, and fertilised separately with milt from two randomly selected males to create paternal half-sib family groups (n = 26 in year 1, n = 25 in year 2). Half of the eggs from each half-sib family were transported to the traditional hatchery and half were maintained at the stream-side hatchery. Eggs were incubated and hatched in heath trays at both hatcheries.

Dispersing larvae (DL)

Sampling for larval *A. fulvescens* passively dispersing downstream from adult spawning areas has been shown to be a viable collection strategy (Auer & Baker 2002; Smith & King 2005). Larval sampling was conducted during a 5-h period, beginning at dusk (21:00 hr) and ending in the early morning (03:00 hr) at a sampling location ≈2 km downstream (Site 4 in Fig. 1) from the primary spawning areas on the Upper Black River (Fig. 1). Five D-framed drift nets were set across the stream channel to capture dispersing larval sturgeon. This design was replicated nightly each year for the entire drifting period. Nets were checked hourly and the larvae within each net were enumerated. Larval *A. fulvescens* captured each evening were reared at the stream-side hatchery until the end of the drifting period. The larvae were then divided between the two hatchery environments. The minimum numbers of adults contributing to larval drift based on capture data were 154 and 234 for the first and second years, respectively (Crossman 2008).

Naturally produced eggs (NPE)

Systematic kick-net sampling was conducted along transects immediately downstream of observed spawning locations to collect eggs that were naturally fertilised and deposited in the stream substratum.

Transects were conducted perpendicular to the river current and kick-net sampling was conducted for 10 s every one metre across the stream channel. Transects were continued downstream in intervals of 5 m until no eggs were collected in consecutive transects. Eggs were incubated and hatched at the stream-side rearing environment because of pathogen concerns of the traditional hatchery. Larvae were then divided between the two rearing environments.

Fish stocking and assessment

The Upper Black River was used as an experimental release site to test whether hatchery rearing environment method of gamete/larval collection and age were not significant predictors of post stocking survival. The release and assessment area encompassed a stream region of ~4 km (Fig. 1). Three age classes were stocked into the Upper Black River. Fish at 8 and 13 weeks of age were released during 2005 and 17 week old fish were released in 2006. Prior to release, all individuals were measured for total length (TL, cm), weighed (g) and tagged with a coloured implant elastomer dye (Northwest Marine Technology, WA, USA) unique to its rearing environment and gamete/juvenile collection method. Elastomer was injected on the ventral side of the rostrum where colour markings were easily distinguished. Juvenile *A. fulvescens* were transported from the traditional hatchery to the stream-side hatchery 1 day prior to each release. All age classes were acclimated to ambient stream conditions within the stream-side hatchery for >12 h prior to being transported to the release site. All fish were released simultaneously at the same location on the Upper Black River (Fig. 1).

The region of the river was chosen because of increased catchability and ease of replication of methods across the different stocking ages. The release location did not represent a stream region in which large numbers of juvenile sturgeon would naturally be present at the three ages of release. Prior to release, four capture locations were established downstream of the release site (Fig. 1). Within each site, four D-framed drift nets were deployed across the stream at equal intervals. Net position was recorded using a GPS unit. Water velocity (m s^{-1}) and depth were recorded at the mouth and corners of each net. Nets were checked and emptied at hourly intervals following release. Sampling persisted for approximately 24 h following release. Sampling at the most upstream assessment site was discontinued for all release ages after no juvenile sturgeon were captured in consecutive hours. Each captured individual was examined for elastomer marks and TL was recorded. Fish were then released immediately downstream of the drift nets.

Block nets were deployed downstream of the third assessment site for both the 13- and 17-week release ages. During the 17-week release a second block net was deployed below the fourth downstream site. The block nets consisted of leaders and large fyke nets (mesh of both: 0.32 cm^2). Leaders were used to guide dispersing *A. fulvescens* into fyke nets. Block nets were deployed to estimate drift net capture efficiency, and to use as a second gear type to assess downstream dispersal, but were not installed during the 8-week release. Sampling protocols for the block nets were consistent with those for the drift nets.

DC tow-barge electric fishing equipment was used 1 h following the releases to survey the stream predator community to assess sources of mortality for released juvenile *A. fulvescens*. A single pass fishing was started at the release location and proceeded downstream past all assessment sites, with captured predators held in an aerated container until the end of the pass. Gastric lavage (Foster 1977) was used to expel the stomach contents of all captured fish. All predators containing juvenile sturgeon were measured for TL (cm) and gape width and height (cm).

Statistical analysis

All statistical analyses were performed using R (R Development Core Team 2007, http://www.rproject.org). Data were tested for normality and homogeneity of variances using a Shapiro–Wilk test and by examining residuals versus fitted values in R. In cases of non-normality or heterogeneity of variances, analyses were run using arcsine-square-root-transformed survival data and log transformed TL data. For analyses within and among release ages, only data from the drift net captures were included. Two dependant variables, the proportion of fish recaptured and the TL of recaptured fish, were examined. The proportion of fish from each collection method and rearing environment captured was calculated for each hourly sampling interval at each assessment site for all release ages. All recaptured proportion values were weighted by the total number released from each hatchery environment and the different collection methods within each release age. A general linear mixed-effects model was used to examine the effects and interactions of six independent variables on the proportion of fish recaptured. Fixed effects included assessment site of capture, gamete/juvenile collection method, rearing environment, net location, physical stream variables (depth and flow measured at the net), and the age of release. Time of capture was included as a random variable in the model. Interactions among age, rearing environment, time of capture and assessment site were also estimated. Data collected from each block net, used in the releases at 13 and 17 weeks, were analysed separately because of lack of comparability to the drift nets and because it was not possible to replicate the sampling strategy across all release ages. The same model was used for the block net as the drift nets except for the assessment site variable. Efficiency estimates for the drift nets were calculated by dividing the total amount of fish recaptured in the drift nets by the total amount of fish recaptured within the same time period (both gear types).

For all stocking events, the response variable TL of recaptured juvenile *A. fulvescens* was analysed using a general linear mixed effects model. In this model, the response was the observation of TL for a specific fish at a distinct sampling time and at a specific sampling site. Fixed effects included capture site, collection method and rearing environment. Time of capture was included as a random effect in the model. The interaction effects between all independent variables were also examined. The independent variable age was not examined in the analysis of TL because there were significant differences between ages in terms of TL at the time of release. The block net data were analysed using the same models.

Mean differences in the TL of recaptured fish in relation to the TLs at the time of release for each treatment group at each release age were tested using a two-factor ANOVA. The response variable (TL) was predicted by period (prior to release or recapture), time (recapture hour), physical stream variables, net location and the interaction between the independent variables. Analyses were conducted separately for fish captured in the block nets and drift nets for each release age. Stream depth and flow, and net position were removed from the final model because of they were insignificant. To test for biases in size-based catchability between the two gear types, an unpaired t-test was used to test for mean differences in the TLs of recaptured fish from each gear type. This was conducted separately for data from the 13 and 17 week releases.

Results

Although sampling was conducted for 24 h, no juvenile *A. fulvescens* was captured beyond 15 h after release. Physical conditions, including water depth and velocity were not statistical predictors of capture rate, but varied between the release ages (Table 1).

Variation in recapture rates

Based on 11 721 juvenile *A. fulvescens* released across each collection method, hatchery rearing environment and age class (Table 1), significantly higher recapture rates were found for fish released at 17 weeks (15%) of age ($F_{2,125} = 6.87$, $P < 0.01$) than fish released at 8 (5%) and 13 weeks (10%) of age (Fig. 2). During the 8 and 13-week releases, a significant main effect of hatchery rearing environment on the proportion of fish recaptured was observed. Significantly fewer recaptured fish ($F_{3,125} = 10.43$, $P < 0.01$) were reared in the traditional hatchery environment relative to the stream-side hatchery (Fig. 2). Significantly fewer fish were captured at increasing downstream distances (2 and 4 km) from the release site ($F_{8,125} = 2.23$, $P = 0.03$) relative to the most upstream assessment sites (0.5 and 1 km) for all release ages (Fig. 2). Significantly more fish were recaptured at downstream assessments sites in the 13-week release than the 8-week release and in the 17-week release compared with both the 8 and 13 week releases (Fig. 2). There was a significant effect of time following release on the proportion of fish recaptured ($F_{23,125} = 4.99$, $P < 0.01$). Fish moving downstream rapidly after release at 8 and 13 weeks of age were mostly captured during daylight hours, but at 17-weeks of age, few fish were captured during the daylight hours immediately following the release (Fig. 3). A large proportion of total captures occurred during evening hours >8 h

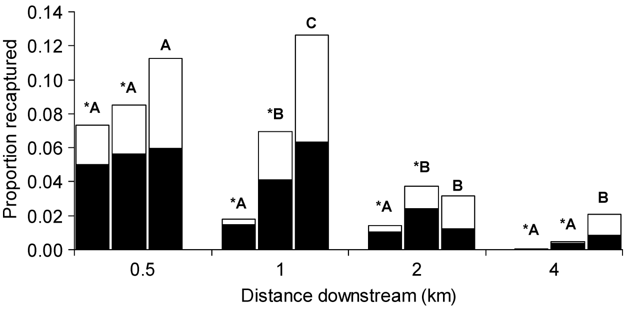

Figure 2. The proportion of juvenile lake sturgeon recaptured at downstream assessment sites (Sites 1–4; Fig. 1) that were reared in different hatchery environments. Ages 8, 13, and 17 weeks are represented within a downstream interval respectively reading left to right. Asterisks represent significantly greater proportions of recaptured fish that were reared at the stream-side hatchery versus the traditional hatchery. Letters (A, B, C) correspond to statistically greater proportions of fish recaptured at the different release ages.

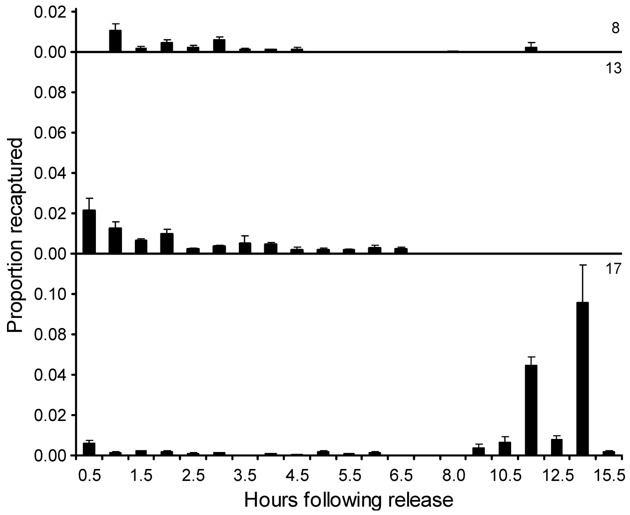

Figure 3. The proportion of juvenile lake sturgeon (means ± 1 SE) captured at consecutive hours following release into the Upper Black River at 8, 13, and 17 weeks of age.

following release (Fig. 3). There was no main effect of collection method on recapture success among release ages. However, differences among collection methods (Table 1) occurred within assessment sites at distinct time periods and although no overall trends emerged from the analysis these results are important given differences in measures of genetic diversity between collections methods found by Crossman (2008). For example, fish released from the stream-side DGT method were recaptured in significantly higher proportions ($F_{4,152}$ = 2.58, P = 0.04) than other methods. At 13 weeks, juvenile *A. fulvescens* from the DL method were recovered in significantly lower proportions ($F_{4,152}$ = 2.79, P = 0.03). Finally, NPE were represented in significantly higher ($F_{4,152}$ = 3.54, P < 0.01) proportions of recaptured juveniles at the oldest release age.

Within the 13-week stocking event, the block net below the third downstream site recaptured 10.3% of the total number of fish released. There were significantly more fish moving downstream of this assessment site than from the stream-side hatchery environment ($F_{1,27}$ = 3.04, P < 0.01). Efficiency of the drift nets varied hourly with a mean (±1 SE) of 12.6 ± 2.8% during the 13-week release. There were no distinct trends in treatment differences among sampling times. Within the 17-week stocking event, the block net below the third downstream sampling location recaptured 15.0% of the total number of fish released. Drift net efficiency was 14.3 ± 5.2%. There was no effect of rearing environment on the proportion of fish moving through this site at any time during the sampling period. As with the drift nets for this age class, significantly more fish were captured at later sampling hours ($F_{7,11}$ = 38.52, P < 0.01). The block net below the fourth downstream site recaptured 19.3% of the fish released. Drift net efficiency was 6.2 ± 2.1%. There was no effect of rearing environment on the proportion of fish recaptured. Again, significantly more fish were caught at later sampling hours than hours immediately following release at this downstream site ($F_{6,8}$ = 11.82, P < 0.01). Although the drift net efficiency varied hourly, results from the block nets were consistent with those from the drift nets.

Variation in size

Documenting differences in TL within and among release ages is important for determining size-based downstream dispersal and as a factor affecting differential survival. No significant differences were observed between rearing environments in the TL of fish prior to release for the 8 and 13-week release ages (Table 1). At 17 weeks of age fish from the traditional rearing environment were significantly larger ($F_{1,609}$ = 11.52, P < 0.01) than those reared at the stream-side hatchery at the time of release (Table 1). There was no main effect of rearing environment or treatment type on the size of recaptured fish in both the 8- and 13-week releases. There was an effect of rearing environment in the 17-week release with recaptured fish from the traditional hatchery significantly larger than those from the stream-side hatchery ($F_{26,583}$ = 22.32, P < 0.01). In all release ages there was a significant positive effect of time following release on the size of recaptured fish (Fig. 4). Fish captured in later post-release were significantly larger than

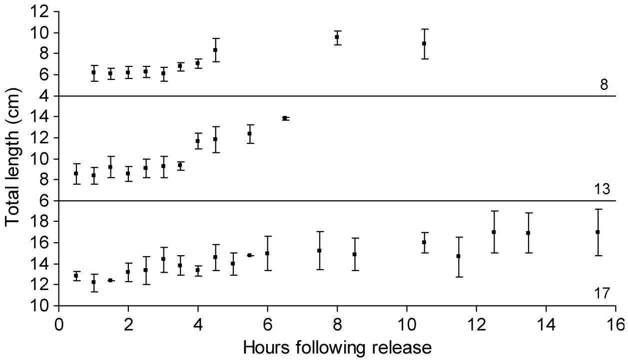

Figure 4. Total length (means ± 1 SE) of recaptured juvenile lake sturgeon over time following release at 8, 13, and 17 weeks of age into the Upper Black River.

fish captured soon after release in the 8-week ($F_{17,202}$ = 4.06, $P < 0.01$), 13-week ($F_{18,302}$ = 7.46, $P < 0.01$), and 17-week ($F_{26,583}$ = 22.32, $P < 0.01$) releases (Fig. 4).

Sizes of fish captured in the block nets at both 13 and 17 weeks revealed trends consistent with those from the drift nets. There was a significant increase in the TL of fish captured in successive sampling periods post-release in the block during the 13-week release. The same results were found for both block nets during the 17-week release ($F_{7,621}$ = 23.97, $P < 0.001$).

During the release at 8 weeks of age, the mean TL of recaptured fish was significantly smaller ($F_{5,589}$ = 11.50, $P < 0.01$) than the mean TL prior to release with the exception of the final recapture period. This was consistent in the 13-week release with fish TLs prior to release being significantly larger than recaptured individuals at all hours except the final net check ($F_{6,1596}$ = 65.82, $P < 0.01$). The mean TL of fish captured in the block net at 13 weeks of age was significantly smaller than mean TL prior to release for all hourly checks ($F_{7,1597}$ = 56.91, $P < 0.01$). At 17 weeks of age, juvenile *A. fulvescens* recaptured in the drift nets were significantly smaller on average than prior to release for all hourly checks except the final recapture period ($F_{4,1191}$ = 7.07, $P < 0.01$). Fish recaptured in the block nets at 17 weeks of age were significantly smaller for all recaptured periods except the final two net checks for both the third ($F_{6,916}$ = 16.53, $P < 0.01$) and fourth most ($F_{6,2859}$ = 17.59, $P < 0.01$) downstream sites. There was no significant difference in the size of juvenile sturgeon recaptured in the block net compared with the drift nets (d.f. = 392, t = 1.293, P = 0.197) during the 13-week release. During the 17-week release, there was no significant difference in sizes between the gear types at the third most downstream site (d.f. = 691, t = 0.092, P = 0.93), but a significant difference was found at the fourth most downstream assessment site (d.f. = 848, t = 3.51, $P < 0.01$) with fish in the block and drift nets averaging 153.47 ± 0.97 mm and 138.35 ± 3.80 mm, respectively.

Evidence of mortality

The sources or rates of mortality by fish predators following release could not be quantified using the tow barge electric fishing gear. During the release at 8 weeks of age, two rock bass, *Ambloplites rupestris* (Rafinesque) were found to have juvenile *A. fulvescens* in their stomachs. Further attempts to quantify predation at later release ages proved unsuccessful. At both 8 and 13 weeks of age, rusty crayfish, *Orconestes rusticus* (Girard) were observed preying upon juvenile *A. fulvescens* after release.

Discussion

Sampling protocols providing quantifiable and comparable results are currently lacking for age-0 *A. fulvescens* (Holey *et al.* 2000), despite recent advances in visual surveys (Benson *et al.* 2005). Using this natural stream as an experimental tool was useful to quantify survival and downstream movement for a species that is otherwise difficult to capture and enumerate. Assessments of juvenile *A. fulvescens* released at 8, 13 and 17 weeks of age found low rates of recovery and inferentially high rates of mortality. Rates of recovery increased with increasing age. The assumption that low rates of recovery lead to low survival is important given the extension of the results towards developing both hatchery and stocking programmes. Visual observations combined with the results of this study demonstrated that mortality was occurring following release. The use of recovery rates as direct measures of survival should be undertaken with caution, but this approach is practical given the susceptibility of juvenile *A. fulvescens* to mortality by a number of different predators (Crossman 2008).

An effect of hatchery rearing environment on juvenile *A. fulvescens* survival and movements was found when juveniles were stocked at 8 and 13 weeks of age. These differences were based on the large number of individuals released (n = 11 721) and is an indication that even moderate differences in hatchery rearing environments potentially related to domestication (e.g. Lynch & O'Hely 2001) can play an important role in post release movements and survival. The effects of rearing environment immediately following release may be reduced by releasing fish at older ages as shown from the lack of differences attributed to hatchery environment for 17-week old fish. Crossman (2008) documented no difference in overwinter survival for juvenile *A. fulvescens* reared in different hatchery environments and released at 6 months of age, supporting the observation that the effects of rearing environment may diminish with age. Estimates of survival for hatchery produced juvenile white sturgeon, *Acipenser transmontanus* (Richardson), in the first year following release can be as high as 84%, with substantial variation surrounding annual estimates attributed to density dependant factors (Ireland *et al.* 2002; Justice *et al.* 2009). However, theoretical (Lynch & O'Hely 2001) and empirical (Araki *et al.* 2007) data on the effects of hatchery rearing environment on reproductive success of hatchery-reared fish dictates that further research be conducted for sturgeon at or beyond the time fish sexually mature.

Significant differences in recapture rates between different hatchery environments indicate the need to tailor culture methods to maximise probabilities of survival through important life-history transitions. This is especially important when dealing with threatened or endangered species such as *A. fulvescens*. Modifying juvenile rearing environments to approximate natural conditions is increasingly used to minimise the degree of domestication for captive animals (Flagg & Nash 1999). Studies of fish reared in different environments have documented differences in behaviour (Olla *et al.* 1998; Berejikian *et al.* 1999; Flagg & Nash 1999), post stocking survival (Wiley *et al.* 1993; Maynard *et al.* 1996), growth rates (Mesick 1988), social rankings (Berejikian *et al.* 2000) and development of appropriate body camouflage colouration (Maynard *et al.* 1996). Olla *et al.* (1998) suggested that fish reared in a sensory limited hatchery environment have poor feeding efficiency and are less capable of avoiding predators. Differences between the stream-side hatchery and the traditional hatchery used in this study were likely of a sensory nature because physical rearing conditions such as tank size, shape, flow rate and feeding regimes were consistent between the two environments. Juvenile *A. fulvescens* reared in the stream-side hatchery may have benefited from fluctuations in temperature mimicking natural environmental conditions. Mechanical filtration within the stream-side hatchery, although effective at removing larger particulates, likely allowed exposure to stream organic material and biological organisms. Furthermore, the water at the stream-side hatchery potentially contained natural stream chemical cues from either conspecifics, predators or other species.

A significant effect of time on fish capture was observed. During releases conducted at 8 and 13 weeks of age, fish were captured at significantly earlier times following release relative to recoveries at 17 weeks of age (Fig. 3). Fish released at 17 weeks dispersed downstream after sunset. This negative photo-tactic or nocturnal behaviour has been noted with *A. fulvescens* larvae dispersing downstream of spawning grounds (Auer & Baker 2002) and similar behavioural trends have been noted in field (Chiasson *et al.* 1997; Benson *et al.* 2005) and hatchery studies on juveniles (Peake 1999). Such nocturnal behaviour has been found in other sturgeon species (Kynard & Parker 2005; Kynard *et al.* 2005) and fish species (Crisp 1991; Bradford & Taylor 1997), and is a dominant feature of migration and foraging in the first year of life. Variability in the proportion of juveniles recaptured was high during the early hours following release at the 8 and 13-week age classes. This is probably because most individuals moving passively in the current were unable to control their rate of downstream dispersal. A notable decrease was observed in the variability in sizes of fish dispersing downstream later in the day at 8 and 13 weeks, and this can be attributed to the movement of primarily larger fish (Fig. 3). There was also a significant effect of fish size on dispersal at 17 weeks of age, but the variation surrounding the estimates was noted in later hours, opposite to that of the earlier age classes (Fig. 3). This increase in variation at later time periods following release indicates that the majority of juveniles had an ability to choose dispersal time. Size-based downstream dispersal has been noted with other fish species. Bradford and Taylor (1997) found that larger Chinook salmon, *Oncorhynchus tshawytscha* (Walbaum), fry immediately post emergence had a higher probability of dispersal during night-time hours than smaller individuals of the same cohort. Age-dependant differences in physical abilities, as suggested by differences in timing and size distributions of dispersing juveniles, should be used to develop release strategies for juvenile *A. fulvescens*.

Lack of a significant main effect of collection treatment on recapture, timing of dispersal or size at recapture suggests the source of gametes of larvae was not as important as rearing environment or age at release. However, differences in recapture rates among individuals from different collection methods across three ages of release for juvenile *A. fulvescens* were found. The variability between individuals from the different methods is important given Crossman (2008) found differences in mean co-ancestry (inter-relatedness of progeny) among juveniles from different collection methods and rearing environments. Juveniles from direct gamete had higher levels of co-ancestry relative to naturally produced eggs and dispersing larvae because of contributions from comparatively fewer adults and comparatively higher inter-family variance in egg survival (Crossman 2008). If probabilities of survival are non-independent relative to family (Crossman 2008), or if body size is heritable, then differences in survival and size at recapture would be expected among collection methods given the large number of individuals released. Further studies, including use of genetic determination of parentage to document family-specific bias in survival and size following release would help resolve questions of the relative effects of environmental and genetic factors affecting survival in sturgeon.

Estimates of recapture success and stream movements among fish from different collection method within each release age are robust because of the large number of fish released (Table 1). However, comparisons of results across age classes should be interpreted in the context of limitations imposed by the sampling design. Catchability increases with increases in body size of fish (Borgstroem & Skaala 1993), and models that assume equal catchability typically underestimate the actual number of fish in the population (Mäntyniemi *et al.* 2005). Juvenile *A. fulvescens* were stocked over 2 years because of a combination of rearing limitations and the need for large sample sizes for release. The use of the block nets as a means of assessing the drift net efficiency and as second gear type was important. Consistency of results between the two gear types was encouraging because drift net efficiency was generally low. Furthermore, only differences in size of fish between the two gear types at one assessment site were documented. This was at the most downstream assessment site where slower water velocities (Table 1) likely allowed the largest juvenile *A fulvescens* to elude the drift nets but not the block nets. Finally, transport of juveniles reared at the traditional hatchery to the stream-side hatchery may have posed increased levels of stress. However, transportation-related effects can be considered as part of the traditional hatchery treatment, as fish transport often occurs over long distances and fish are routinely released without any period of acclimation. Fish used in the present study were acclimated at the stream-side hatchery overnight (> 12 h) prior to release to decrease the likelihood of transportation-related effects.

River conditions varied between release events with the lowest water velocities occurring during the 17-week release. Lower drift net efficiency at the lowest assessment site at 17 weeks of age can likely be attributed to the decrease in water velocities. Studies have indicated that higher water velocities can strongly influence downstream displacement of young fish (Daufresne *et al.* 2005). This may have been the case for juvenile *A. fulvescens* at 8 and 13 weeks of age. However, a subset of larger fish were found that were able to control their rate of dispersal in each release.

It is imperative that well-defined, species-specific protocols be developed for stocking programmes (Cowx 1999). Supplementation protocols should be tailored to species specific ecologies and behaviours, and assessments should be made to quantify estimates of survival following release. Protocols have been developed for many recreationally and commercially important species that incorporate ecologically important information. Largemouth bass, *Micropterus salmoides* (Lacepède) has low dispersal following release, indicating that localised supplemental stocking in distinct areas of complex reservoirs may increase benefits at the population level (Copeland & Noble 1994). The matching of stocking times and locations to appropriate food resources is important to walleye, *Sander vitreum (*Mitchill*)* introductions (Ellison & Franzin 1992). The present results also indicate that supplementation strategies should focus on night releases and in areas of moderate flow. These variables ensure that rates of dispersal are high immediately following release and will likely have an indirect effect on levels of predation by reducing mortality incurred by visual predators. It is critical that release sites are evaluated on a system by system basis, with emphasis placed on determining areas that maximise suitable juvenile nursery habitat. For *A. fulvescens*, natural juvenile rearing habitat has been defined as predominantly shallow riverine habitat consisting mostly of sand (Chiasson *et al.* 1997; Benson *et al.* 2005) or smaller gravels (Holtgren & Auer 2004).

To conclude, empirical and experimentally rigorous evaluations of hatchery-rearing and stocking programmes for little studied species such as *A. fulvescens* is essential. Stream-side rearing may be advantageous to small/young *A. fulvescens* by exposing them to an enriched environment prior to release. However, there may be an age/size threshold at which the effects of rearing environment on movements and survival immediately following release are diminished. Ways in which progeny are collected for hatchery programmes will depend on access to life stages but efforts should be made to maintain adequate levels of genetic diversity given the variability found in recapture rates between collection methods used in this study. Further work that characterises mechanisms influencing differences among hatchery rearing environments for *A. fulvescens* is important to the development of system specific management prescriptions.

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