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Evaluation of relocation of unionid mussels into artificial ponds

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Abstract. Relocation of unionid mussels into refuges (e.g., hatchery ponds) has been suggested as a management tool to protect these animals from the threat of zebra mussel (*Dreissena polymorpha*) invasion. To evaluate the efficacy of relocation, we experimentally relocated 768 mussels, representing 5 species (*Leptodea fragilis, Obliquaria reflexa, Fusconaia flava, Amblema plicata,* and *Quadrula quadrula*) into an earthen pond at a National Fish Hatchery or back into the river. In both locations, mussels were placed into 1 of 4 treatments (mesh bags, corrals, and buried or suspended substrate-filled trays). Mussels were examined annually for survival, growth (shell length and wet mass), and physiological condition (glycogen concentration in foot and mantle and tissue condition index) for 36 mo in the pond or 40 mo in the river. We observed significant differences in mortality rates between locations (mortality was 4 times greater in the pond than in the river), among treatments (lowest mortality in the suspended trays), and among species (lower mortality in the amblemines than lampsilines). Overall survival in both locations averaged 80% the 1st year; survival in the pond decreased dramatically after that. Although length and weight varied between locations and over time, these changes were small, suggesting that their utility as short-term measures of well being in long-lived unionids is questionable. Mussels relocated to the pond were in poor physiological condition relative to those in the river, but the magnitude of these differences was small compared to the inherent variability in physiological condition of reference mussels. These data suggest that relocation of unionids into artificial ponds is a high-risk conservation strategy; alternatives such as introduction of infected host fish, identification of mussel beds at greatest risk from zebra mussels, and a critical, large-scale assessment of the factors contributing to their decline should be explored.

Key words: unionids, relocation, conservation, survival, growth, physiological condition.

Unionid mussels are the most imperiled group of animals in the United States (Master et al. 1998). About 70% of the species in this group are either extinct, endangered, threatened, or listed as species of special concern (Williams et al. 1993). Consequently, many federal, state, and provincial agencies are examining approaches to conserve this declining fauna. Relocation of unionids is presently being used extensively as both a conservation and management tool (Cope and Waller 1995). Relocations have been conducted for a variety of reasons, including recolonization of areas where populations have been decimated by pollution (Sheehan et al. 1989), removal of mussels from construction activities (Dunn 1993, Trdan and Hoeh 1993), protection from thermal discharges downstream of hydroelectric dams (Heinricher and Layzer 1999), protection from harvest pressure (Obermeyer 1997), and protection from colonization by the zebra mussel *Dreissena polymorpha* (Sickel et al. 1997).

Relocation projects have met with limited success. Cope and Waller (1995), who reviewed 33 relocation projects that moved 90,000 mussels, reported a mean survival of 50%, suggesting that existing relocation methods have been

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stressful to mussels. Furthermore, most relocations were monitored for <1 y, and <20% were monitored for 5 y. The success of most relocation projects has been predominately judged by survival; few studies examined other measures such as growth, recruitment, or condition. In addition to low survival, relocation raises serious ecological and evolutionary concerns, particularly because most mussels are moved with little attention to genetic or disease issues (Villella et al. 1998).

There is no accepted procedure for relocating mussels, and it is not surprising that the results are highly variable. Thus, it is extremely difficult to draw conclusions about the usefulness of this management tool. For example, Dunn (1993) reported that 50% of the mortality occurs during the 1st few months after relocation, whereas Layzer and Gordon (1993) reported high survival (>75%) for the first 2 y after relocation but low survival (21%) 4 y after relocation. If the latter is true, studies that monitor mussels for only a few years could overestimate the success of relocations. For these reasons, along with the long life span of many unionids (up to 100 y), we suggest that measures other than survival be used to judge the success of relocation efforts and to ascertain the physiological health of unionids during relocation events. The physiological condition of unionids may decline long before mortality occurs (Haag et al. 1993).

Glycogen is the principal storage form of carbohydrates in many aquatic invertebrates (De Zwaan and Zandee 1972, Hummel et al. 1989). Glycogen concentration has been used as an indicator of physiological condition in unionids to assess stress from contaminant exposure (Hemelraad et al. 1990), zebra-mussel infestation (Haag et al. 1993), and relocation (Naimo and Monroe 1999). For example, Naimo and Monroe (1999) observed that glycogen concentrations in *Amblema plicata* declined 80% in mantle tissue and 56% in foot tissue 24 mo after relocation from the Upper Mississippi River into an artificial pond. Similarly, the tissue condition index (TCI, the ratio of dry tissue mass to dry shell mass) has been used as an indicator of the relative health of unionids after exposure to contaminants (Naimo et al. 1992) and infestation by zebra mussels (Baker and Hornbach 1997).

Proliferation of zebra mussels has prompted biologists to identify factors that may influence the success of mussel relocations. We were asked by the US Fish and Wildlife Service to evaluate the success of relocation into a hatchery pond because the infrastructure for this management approach already exists throughout much of the United States. Our objectives were 1) to determine if survival, growth, and physiological condition differed between unionids relocated to an artificial pond and mussels held under similar conditions in the Upper Mississippi River, 2) to assess species differences in the survival of mussels, and 3) to assess which method of holding mussels yielded the best survival, growth, and physiological condition.

Methods

Experimental design

We removed mussels from the Upper Mississippi River, quarantined them for 35 d to reduce the likelihood of transporting zebra mussel veligers, and relocated them into an artificial pond or put them back into the river. We placed mussels into 1 of 4 holding structures (treatments) in both locations to facilitate recovery. Each treatment was replicated 4 times for a total of 16 experimental units per location. The treatments included 1) mesh bags—nylon bags (51 × 64 cm, mesh size 2.5 cm) containing 24 pockets that were suspended ~5 cm above the sediment–water interface, 2) suspended, substrate-filled trays (suspended trays)—1 m^2 metal baskets in a metal framework that were placed 45 to 60 cm above the sediment–water interface, 3) buried, substrate-filled trays (buried trays)—similar to treatment 2 except these structures were placed at the sediment–water interface, and 4) corrals—a metal barrier enclosing 1 m^2 of natural substrate. Suspended and buried trays were filled with dredged material from the river that was deposited before the initial appearance of zebra mussels. All experimental units in the pond were covered with chicken wire to reduce predation by furbearing mammals.

The dredged sediments and the top 4 to 6 cm of naturally occurring sediments in the river and pond were characterized for organic C by loss on ignition (APHA et al. 1995) and particle size by the sieve-pipet method (Guy 1969, Plumb 1981). The mean (±1 SE) % organic C was 0.3 ± 0.01% in the dredged sediments, 2.0 ± 0.02% in the river sediments, and 7.3 ± 0.01%

in the pond sediments. Particle size also varied substantially among the 3 sediments. Sediments from the river were 76% sand and 24% silt and clay. In contrast, sediments from the pond were 23% sand and 77% silt and clay. The dredged material was almost exclusively sand (99%).

Mussel collection and quarantine

In February 1995, we obtained ~60 mussels from River Mile 673 of the Upper Mississippi River near Victory, Wisconsin, and transported them to the US Fish and Wildlife Service's Fish Disease Control Center in Onalaska, Wisconsin. These mussels were certified free of bacterial and viral agents. During 1 to 3 May and 18 May 1995, we obtained 1392 mussels of 5 species (160 *Leptodea fragilis*, 205 *Obliquaria reflexa*, 165 *Fusconaia flava*, 663 *Amblema plicata*, and 199 *Quadrula quadrula*) from River Mile 673. Mussels were moved in the spring, before the major spawning period of zebra mussels, to reduce the probability of relocating veligers and because moving mussels early in the season may provide energetic benefits later in the season (Jokela 1996). Each unionid was scrubbed free of zebra mussels during collection. The mean (±1 SD) number of zebra mussels per unionid (averaged over all species) was 0.5 ± 0.3 and ranged from 0 to 13. Unionids were transported after processing to a quarantine pond in coolers lined with ice and layered with burlap bags soaked in well water.

We quarantined all mussels for 35 d in a 0.04-ha concrete pond at the Upper Midwest Environmental Sciences Center in La Crosse, Wisconsin, that contained no inflow or outflow. Dissolved oxygen levels in the quarantine pond were maintained with an electric aerator. Temperature ranged from 13.0 to 26.7°C (mean 18.2°C), dissolved oxygen from 6.3 to 19.8 mg/L (mean 10.8 mg/L), and pH from 7.8 to 10.6 (mean 8.9). All unionids were placed into 1 of 8 net bags (1.2 × 1.8 × 1.2 m deep, mesh size 1 cm). During the quarantine period, 187 mussels died. Survival was 52% in *L. fragilis*, 80% in *O. reflexa*, 85% in *F. flava*, 94% in *A. plicata*, and 96% in *Q. quadrula*. After the quarantine period, mussels were visually inspected for zebra mussels with 2× magnification, their shell lengths and wet masses were measured, and each mussel was tagged with a unique identification number. Two numerically labeled shellfish tags (Hallprint International, Australia) were affixed to the shell of each mussel with cyanoacrylate glue. Each mussel was emersed for only ~5 min during processing. We found no zebra mussels on any live unionid after the quarantine period.

Relocation

In June 1995, we relocated 768 mussels—384 mussels went into an 0.10-ha earthen pond (34 × 24 × 1 m deep, supplied with well water and aerated) at a National Fish Hatchery in Genoa, Wisconsin, and 384 mussels went back into the Upper Mississippi River (River Mile 673). The 16 experimental units were randomly placed in a 4 × 4 array in the pond or river. In both locations, 24 mussels were randomly placed into each of the 16 experimental units: 5 *Q. quadrula*, 5 *O. reflexa*, 3 *F. flava*, 8 *A. plicata*, and 3 *L. fragilis*. These densities approximated the natural densities of mussels in this portion of the river (K. I. Welke, unpublished data). We did not determine the sex or gravidity of the relocated mussels. There were no statistical differences in the shell length or wet mass of mussels between locations or among treatments (ANOVA, all p values >0.05). The grand means (±1 SE) for length (mm) and wet mass (g), respectively, were: *A. plicata*—79.0 ± 0.8, 160 ± 4; *F. flava*—58.3 ± 0.9, 92 ± 4; *L. fragilis*—90.6 ± 2.1, 77 ± 5; *O. reflexa*—45.9 ± 0.4, 39 ± 1; and *Q. quadrula*—73.0 ± 1.1, 143 ± 5.

We recorded survival, shell length, and wet mass of all mussels in September 1995, 1996, 1997, and 1998 (the last monitoring date in the pond was May 1998 because of construction activities). We examined mussels in September to minimize the temperature differential between air and water (Dunn and Sietman 1997) and because few species were brooding glochidia (Watters 1994). In the river, we removed and counted the number of attached zebra mussels annually. In the pond, we suspended one zebra mussel sampler (4 vertically placed PVC plates that were 232, 413, 645, and 930 cm², separated by a 2.5-cm spacer) each May near the experimental units to monitor for veliger settlement. The sampler was checked each September.

Water quality was measured weekly in the pond and biweekly in the river from April to September. We measured dissolved oxygen and temperature (Yellow Springs Instrument Company, Model 57) and pH (Orion, Model 230A)

from April 1995 until April (pond) or September (river) 1998. We measured total alkalinity and hardness monthly at each location (APHA et al. 1995). We also measured chlorophyll *a* concentrations at both locations (APHA et al. 1995) beginning in May 1996. All samples were taken at 0.3 m below the surface in an area near the mussels. The temperature, dissolved oxygen, and pH of surface waters were similar in the pond and the river during the relocation, whereas alkalinity, hardness, and chlorophyll *a* concentrations differed between the pond and river (Fig. 1).

Physiological analysis

Each September, we randomly removed one *A. plicata* (80–90 mm in shell length) from each experimental unit in each location for determination of glycogen concentration and TCI. We also sampled 10 *A. plicata* that were not associated with the relocation (referred to as reference mussels; 80–90 mm in shell length) from River Mile 673 each September to estimate baseline glycogen and TCI. We quickly removed ~100 mg of foot tissue and 50 mg of mantle tissue from near the ventral shell margin in the field (Naimo et al. 1998). Tissues were placed on dry ice and later stored at −84°C until analyzed. Glycogen concentration (mg/g dry mass) was measured following Naimo et al. (1998). The TCI was determined by dividing the dry tissue mass by the dry shell mass and multiplying the quotient by 100 (Naimo et al. 1992). No individual contained >3 zebra mussels.

Statistical analysis

Survival.—Our survival data consisted of measurements of elapsed times until death of individual mussels or the end of the study. Analysis based on the elapsed times until death is preferable to analyses of the proportions of individuals that died because the elapsed times until death contain more information. Analyses of event times can also detect effects better than analyses of the proportions of individuals that experienced those events (Waller et al. 1999). A key feature of such data is that some times-to-death may have exceeded the observation duration. For these instances, we know only that the unknown times to death exceeded some observation duration, and they are said to be right censored. Similarly, some mussels were never again found, alive or dead, after an observation; their times to death are also right censored at the time they were last observed to be alive. Observations at annual intervals, such as in this study, are said to be both left and right (interval) censored; that is, we know only that a death occurred within a 1-y interval. However, to avoid specification of a particular distribution for survival times, which is required for interval censoring, we approximated the times to death as the midpoint of the annual intervals and henceforth assumed only right censoring. We used the semiparametric Cox proportional hazards regression model for right censored data (Cox 1972, Hosmer and Lemeshow 1999) to identify factors that explained the pattern in survival times. This model does not require specification of a particular survival distribution. We arbitrarily selected *Q. quadrula* as the baseline species, corrals as the baseline treatment, and the river as our baseline location; these choices do not affect the overall results. Our full proportional hazards regression model was

$$\lambda_{ijk}(t) = \lambda_0(t) \exp(\zeta_i + \tau_j + \gamma_k) \qquad [1]$$

where $\lambda_{ijk}(t)$ is the hazard function for the ith species in the jth treatment at the kth location at time t; $\lambda_0(t)$ is the corresponding baseline hazard; ζ_i, $i = 1$–4, are 4 parameters for identification of the 5 species; and τ_j and γ_k are effects of treatment and location, respectively. We fitted equation 1 to the survival data by maximizing the partial likelihood, and constructed likelihood-ratio and Wald χ^2 tests for each parameter using the SAS PHREG software (Version 6.12, SAS Institute, Cary, North Carolina). We began with our full regression model (equation 1) and, one-by-one, deleted terms for which the corresponding likelihood-ratio χ^2 test was not significant at the $\alpha = 0.05$ level. Our recorded event times were based on observations at fixed times and consequently contained ties. We used Efron's method to adjust for tied event times (Hertz-Picciotto and Rockhill 1997).

The parameters of the Cox proportional hazards model have natural interpretations that provide informative descriptions of the event times. The hazard function $\lambda(t)$ quantifies the instantaneous mortality rate at time t given survival up to that time. From equation 1, the dimensionless hazard (risk) ratio at time t is given

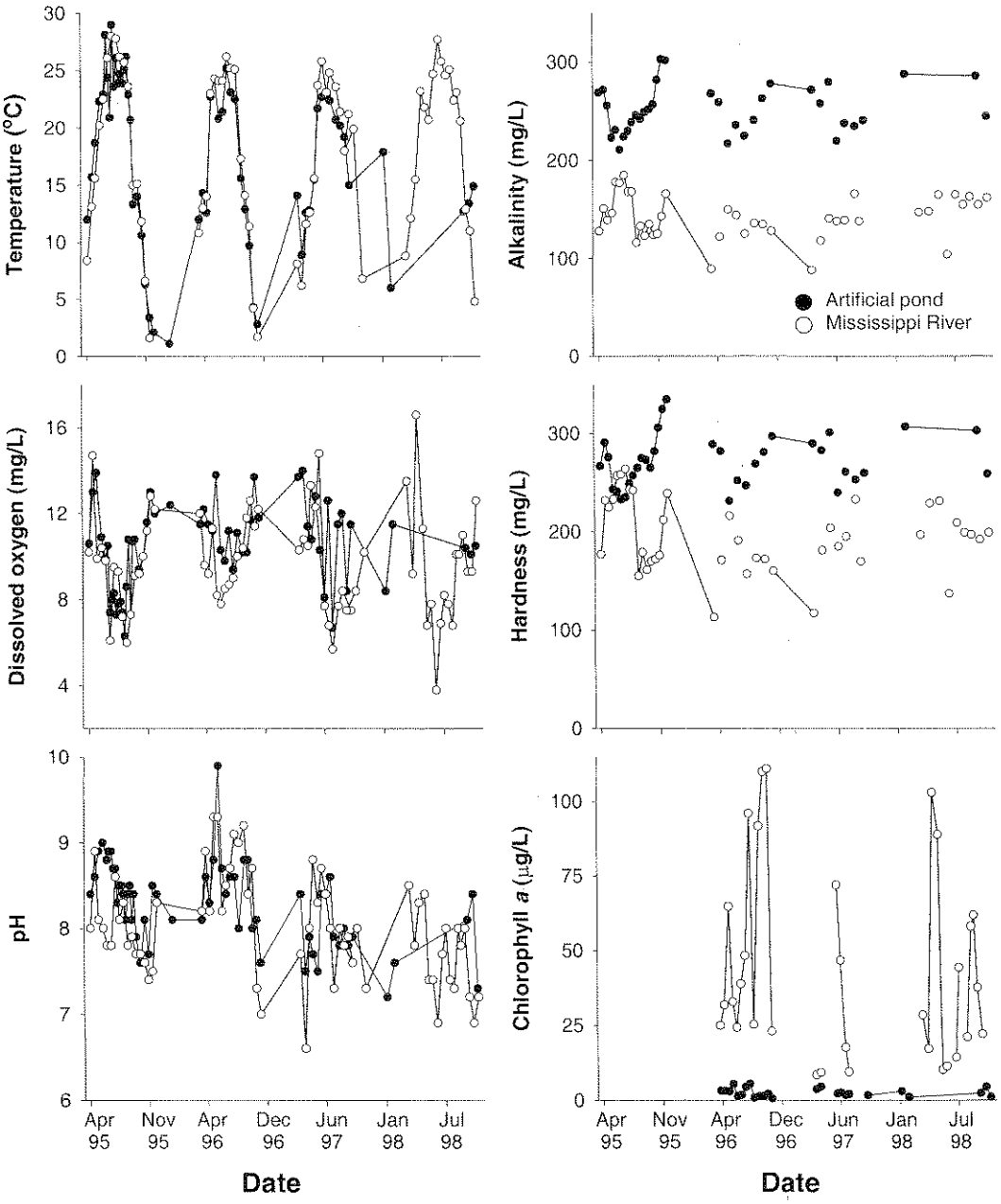

FIG. 1. Temperature, dissolved oxygen, pH, alkalinity (as $CaCO_3$), hardness (as $CaCO_3$), and chlorophyll *a* concentrations of surface waters in the artificial pond and the Upper Mississippi River during the relocation study.

by $HR(t) = \lambda_{ijk}(t)/\lambda_0(t) = \exp[\zeta_i + \tau_j + \gamma_k]$, and quantifies the ratio of the conditional (on survival up to time t) mortality rate for species i in treatment j and location k to the conditional mortality rate for the baseline species, treatment, and location. For categorical variables such as species ζ_i in our analysis, the hazard ratio for species i relative to the baseline species is $\exp(\zeta_i)$. If, for example, $\exp(\zeta_i) = 0.5$, we say that the *relative* hazard or conditional mortality

rate for species i is only 50% of that for the baseline species. By extension, the hazard ratio for species 1 relative to species 2 is $\exp[\zeta_1 - \zeta_2]$. For display of the model results we computed product-limit estimates (Hosmer and Lemeshow 1999) of the probabilities that times until events exceed some time t.

We tested the likely consequences of our assumption that deaths occurred at the midpoints of the annual observation intervals by fitting a fully parametric survival model, based on the Weibull distribution, to the actual interval-censored recordings. The 2 approaches yielded practically identical results, and we preferred to assume deaths occurred at the annual midpoints rather than the Weibull distribution for times to death.

Growth.—Growth of individuals can be quantified by trajectories of body size over time. Each individual generates a growth trajectory, and these trajectories may vary because of effects of treatment, location, and inherent variation among individuals. Our growth data consisted of repeated measurements of shell length and wet mass from individually marked mussels that were randomized to treatment and location. The repeated measurements may be serially correlated within individuals because body size is the result of a cumulative growth history. Further, the duration of this study was short compared with the life spans of these species, and hence we have data on only relatively short segments of the entire growth trajectories from each individual. Asymptotic growth was not observed because of this short duration. Therefore, we modeled growth with a quadratic polynomial in time. We formulated a hierarchical random-coefficients regression model (Davidian and Giltinan 1995) that accommodated all of these features to examine potential differences in individual growth trajectories among treatments and locations. The 1st stage of this hierarchical model describes growth trajectories of individual mussels, and the 2nd stage models variation of these trajectories in response to treatments and locations.

Let $L_k(t)$ denote body size at time t of individual k, k-1. . . 256. Our full 1st-stage model for $L_k(t)$ is given by

$$L_k(t) = \beta_{0,k} + \beta_{1,k}t + \beta_{2,k}t^2 + \epsilon_k(t) \qquad [2]$$

where $\beta_{0,k}, \ldots, \beta_{2,k}$ are parameters that are assumed to be jointly normally distributed. For the kth individual, $\beta_{0,k}$ is the L intercept, $\beta_{1,k}$ is the linear coefficient of t, and $\beta_{2,k}$ is the coefficient for the effect of t^2. For the kth individual mussel, we assumed that the $\epsilon_k(t)$ were normally distributed with mean 0 and variance $\rho^{|\Delta t|}\sigma^2$, where ρ is an autoregressive parameter modeling the serial correlation among measurements separated by the time interval Δt, and σ^2 is a constant portion of the variance. For the 2nd stage, we modeled the response of $\beta_{0,k}, \ldots, \beta_{2,k}$, to the effects of treatment, location, and treatment × location interactions. We fitted this model with the SAS MIXED procedure (Version 6.12, SAS Institute, Cary, North Carolina) and began with a full model containing effects for treatment, location, and treatment × location interactions and deleted terms, one-by-one, that were not significant based on F tests.

Physiology.—We analyzed the physiological data with a generalization of analysis of covariance. Preliminary analyses indicated that the variance in glycogen concentration and the TCI was proportional to the square of the mean. This result is characteristic of the gamma distribution, and we therefore assumed that distribution in a generalized linear model (McCullagh and Nelder 1989). This modeling approach is more flexible and appropriate than use of normal-theory methods on transformed data. Our model for glycogen concentration and TCI is an extension of the analysis of variance that accommodates gamma-distributed responses, and is given by

$$\mu_{jk} = \mu_0 \exp(\tau_j + \gamma_k + \tau\gamma_{jk} + \beta_1 t + \beta_{1,j}t + \beta_{1,k}t + \beta_{1,jk}t + \beta_2 t^2 + \beta_{2,j}t^2 + \beta_{2,k}t^2 + \beta_{2,jk}t^2) \qquad [3]$$

where μ_{jk} denotes mean glycogen concentration or TCI in treatment j and location k, μ_0 is the corresponding baseline mean, τ_j is the effect of treatment j, γ_k is effect of location k, $\tau\gamma_{jk}$ is the treatment × location interaction, β_1 is the coefficient for the linear effect of time t, $\beta_{1,j}$ is the coefficient for the location × time interaction, $\beta_{1,k}$ are 4 coefficients for the treatment × time interaction, $\beta_{1,jk}$ are 4 coefficients for location × treatment × time interaction, and the β_2 coefficients are similar to the β_1 terms but for the quadratic effect of time. We used the SAS GENMOD procedure (Version 6.12, SAS Institute, Cary, North Carolina) to obtain maximum-likelihood

TABLE 1. Percent recovery of 5 species of unionids, whether alive or dead, after a 36-mo relocation into an artificial pond or a 40-mo relocation into the Upper Mississippi River.

Species	Number relocated	Treatment			
		Mesh bags	Buried trays	Suspended trays	Corrals
		Artificial pond			
Amblema plicata	32	100	97	100	94
Fusconaia flava	12	100	100	100	92
Quadrula quadrula	20	100	95	100	100
Leptodea fragilis	12	100	92	100	92
Obliquaria reflexa	20	100	95	100	100
		Upper Mississippi River			
Amblema plicata	32	94	100	94	81
Fusconaia flava	12	92	92	75	50
Quadrula quadrula	20	100	90	90	40
Leptodea fragilis	12	33	75	67	25
Obliquaria reflexa	20	85	85	65	30

estimates of the parameters in the full model, and used the scaled deviance and likelihood-ratio tests to identify unimportant parameters for elimination to produce the simplest model that fit the data. This model is analogous to an analysis of covariance, for normally distributed errors on the log scale, that allows for unequal slopes among combinations of location and treatment for each of 2 covariates.

Results

We recovered 670 live and dead mussels at the end of the study. Recovery of mussels in the pond ranged from 92% to 100%, and was similar among species and treatments (Table 1). In contrast, % recovery of mussels from the river was highly variable, differing substantially among species and treatments (Table 1).

We did not observe any zebra mussels on unionids or the plate samplers in the pond. However, zebra mussels were consistently present in the river, even though each unionid was cleaned annually (Table 2). The numbers of zebra mussels per unionid were consistently lower in the corral treatment than in other treatments (Table 2). The numbers of attached zebra mussels on *A. plicata, F. flava,* and *O. reflexa* were similar, whereas *L. fragilis* and *Q. quadrula* supported more zebra mussels.

Survival

The conditional mortality rates (hazards) of relocated mussels differed significantly between locations ($p < 0.01$). The conditional mortality rate (exponential function of the parameter estimate in the proportional hazards model) in the pond was 4.1 times that in the river. As the experiment evolved, survival was distinctly better in the river than in the pond (Fig. 2). At the end of the experiment, 35% of the mussels that were relocated into the pond were alive, whereas 75% of the mussels relocated into the river survived.

The conditional mortality rates of mussels also differed significantly among treatments ($p < 0.01$). The conditional mortality rates for mussels in suspended trays was only 0.41 times that of mussels in the corrals, 0.36 times that of mussels in the mesh bags, and 0.54 times that of mussels in the buried trays, indicating significantly greater survival ($p < 0.01$) in the suspended trays than in the other 3 treatments (Fig. 2). The conditional mortality rates of mussels in the mesh bags did not differ significantly from those in the corrals, but were significantly greater than those of mussels held in buried trays ($p = 0.01$) or suspended trays ($p < 0.01$). Placement of mussels in the buried trays or in the mesh bags did not result in significantly different mortality rates than did placement in the corrals. At the end of the relocation, survival of mussels in the suspended trays averaged 52% in the pond and 85% in the river (Fig. 2). In contrast, survival of mussels in the other 3 treatments ranged from 20% to 36% in the pond and from 64% to 77% in the river.

We observed species differences in the survival of mussels during the relocation. In particu-

TABLE 2. Mean (±1 SE) number of zebra mussels attached to unionids that were relocated into 4 treatments in the Upper Mississippi River. Mussels were initially relocated in June 1995; zebra mussels were removed and counted each September thereafter. — = no individuals were recorded alive.

Year	*Amblema plicata*	*Fusconaia flava*	*Leptodea fragilis*	*Obliquaria reflexa*	*Quadrula quadrula*
			Mesh bags		
1995	4.1 ± 0.6	3.3 ± 0.9	0	0.8 ± 0.3	3.4 ± 1.0
1996	30.5 ± 3.7	24.1 ± 6.5	17.0 ± 5.1	10.7 ± 1.4	27.9 ± 3.5
1997	19.4 ± 4.1	19.9 ± 7.4	4.5 ± 2.1	4.4 ± 0.8	21.6 ± 3.8
1998	33.0 ± 6.9	46.3 ± 11.5	78.0 ± 19.1	29.6 ± 5.7	75.0 ± 14.8
			Buried trays		
1995	3.7 ± 1.5	1.5 ± 0.7	8.1 ± 3.8	0.7 ± 0.3	19.5 ± 6.8
1996	13.6 ± 3.7	10.5 ± 4.4	18.4 ± 9.5	2.5 ± 0.7	10.6 ± 2.9
1997	71.2 ± 7.8	31.5 ± 5.2	73.3 ± 19.9	37.1 ± 7.3	75.7 ± 13.3
1998	23.6 ± 6.2	10.5 ± 4.9	83.0 ± 50.7	16.8 ± 3.7	30.0 ± 7.5
			Suspended trays		
1995	2.2 ± 0.6	1.2 ± 0.5	0.3 ± 0.2	0.2 ± 0.1	3.3 ± 1.0
1996	14.3 ± 6.8	4.3 ± 1.7	27.1 ± 23.8	6.4 ± 1.9	12.4 ± 2.4
1997	66.7 ± 11.0	50.1 ± 13.1	92.1 ± 13.2	29.3 ± 5.5	93.2 ± 11.4
1998	85.6 ± 16.4	55.4 ± 15.4	34.0 ± 15.9	71.3 ± 10.2	72.8 ± 19.7
			Corrals		
1995	0.5 ± 0.3	0.5 ± 0.2	0.5 ± 0.3	0.2 ± 0.1	0.3 ± 0.1
1996	2.9 ± 0.8	0.4 ± 0.3	—	2.5 ± 1.1	1.3 ± 1.1
1997	23.2 ± 5.1	11.8 ± 3.4	—	6.8 ± 3.0	12.6 ± 5.1
1998	25.8 ± 8.0	21.3 ± 6.8	1.7 ± 0.3	16.9 ± 13.4	37.4 ± 18.1

lar, there were no significant differences in the conditional mortality rates among the 3 amblemines (*A. plicata, F. flava,* and *Q. quadrula*), but the conditional mortality rates of the amblemines were significantly lower than the lampsilines (*L. fragilis* and *O. reflexa*; $p < 0.01$). Within the 2 lampsilines, the conditional mortality rate of *L. fragilis* was significantly greater than that of *O. reflexa* ($p < 0.01$). As a result, the survival probabilities of the amblemines and lampsilines diverged during the course of the experiment, and those of the lampsiline *L. fragilis* became particularly low (Fig. 2). After 36 mo in the pond, survival of the amblemines ranged from 31% in *Q. quadrula* to 46% in *F. flava*; survival of both lampsilines was 27%. In the river, a similar pattern existed with amblemine survival ranging from 78% in *A. plicata* to 82% in *F. flava*, whereas lampsiline survival was only 58% in *L. fragilis* and 71% in *O. reflexa.*

Growth

The individual growth trajectories, as measured by changes in shell length over time, did not differ significantly among treatments for any species. However, the individual growth trajectories differed between locations through the effects of time (time × location) and time^2 (time^2 × location) for each species (all $p < 0.05$) and differed in shape between locations (Fig. 3). Not surprisingly, mussels relocated to the pond initially grew slowly (first 16 mo) and then growth increased in the latter 20 mo of the relocation. In the river, relocated mussels appeared to grow faster during the first 16 mo, followed by slower growth during the last 24 mo. At the end of the study, mussels relocated into the pond averaged <2.8 mm of new shell material in 36 mo, whereas mussels relocated into the river grew between 3.3 (*Q. quadrula*) and 7.1 mm (*L. fragilis*) in 40 mo.

Individual growth trajectories, as measured by changes in wet mass over time, like those for length, did not differ among treatments for any species, but differed between locations through the effects of time (time × location) for each species (all $p < 0.04$) and additionally through the effects of time^2 (time^2 × location) for *A. plicata* and *O. reflexa* (both $p = 0.02$). Therefore, the growth trajectories for *F. flava, L. fragilis,* and *Q. quadrula* were best described by straight lines,

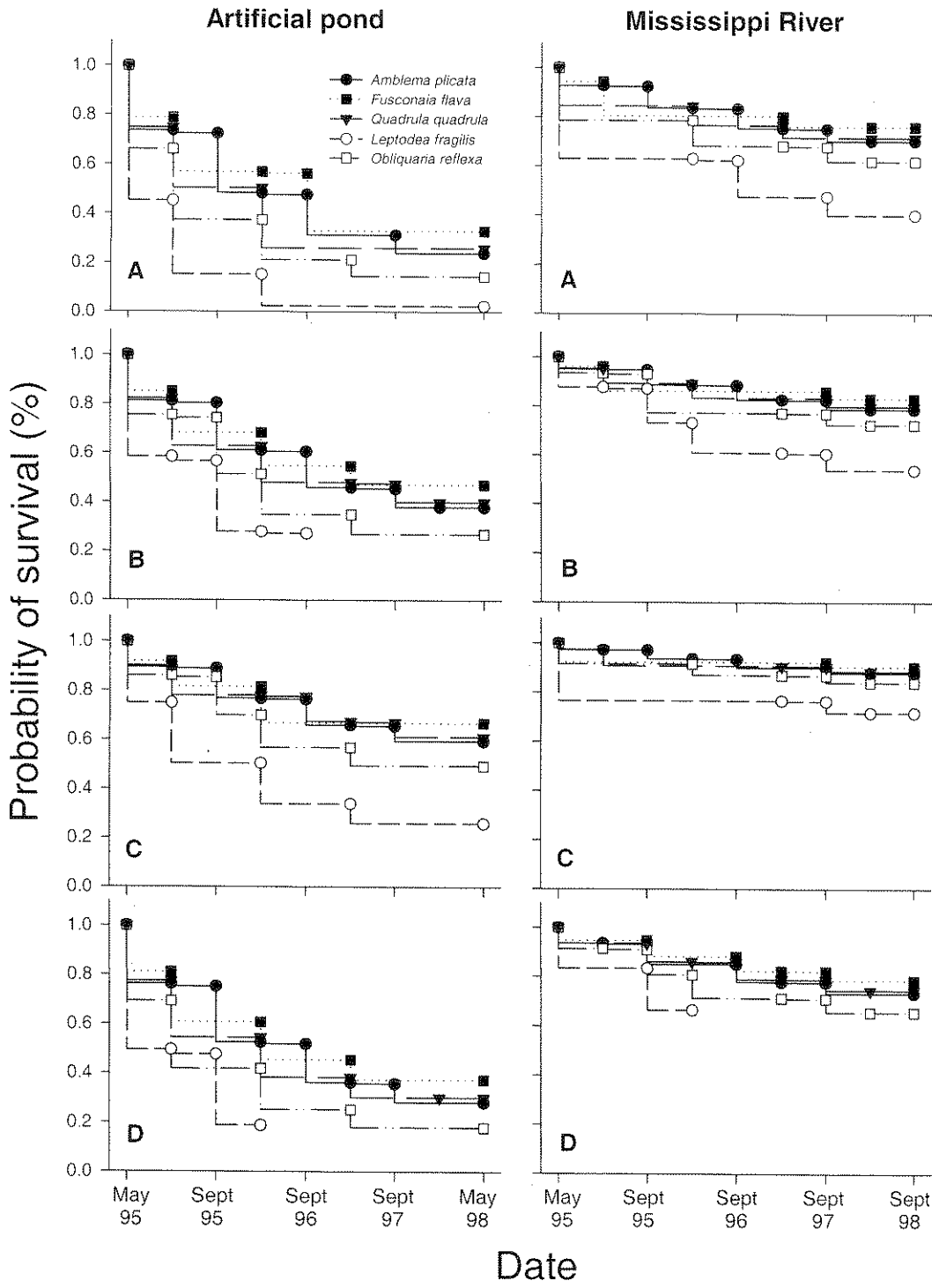

FIG. 2. Survival estimates of 5 mussel species (amblemines—filled symbols, lampsilines—open symbols) during a 36-mo relocation into an artificial pond or a 40-mo relocation into the Upper Mississippi River. Mussels were relocated into 1 of 4 treatments: (A) mesh bags, (B) buried trays, (C) suspended trays, and (D) corrals.

whereas those for *A. plicata* and *O. reflexa* were quadratic functions that differed in shape between locations (Fig. 4). Growth rates, measured by the slopes of the straight lines and gradients of the quadratic curves, were consistently greater in the river than in the pond. Mussels relocated into the pond added an average of 4 g over 36 mo (range: 0 in *Q. quadrula* to 11 g in *L. fragilis*), whereas mussels relocated to the river added an average of 18 g over 40 mo (range: 10 in *O. reflexa* to 25 g in *A. plicata*). For *A. plicata* and *O. reflexa*, the shape of the growth curve (based on mass) was similar to that observed in length; the mussels in the river grew at a faster rate during the 1st part of the study, whereas in the pond, the fastest growth rate was observed in the latter months of the relocation (Fig. 4).

Physiology

During the 35-d quarantine, glycogen concentrations in foot tissue in *A. plicata* declined 44%, from 279 ± 191 mg/g at day 0 to 178 ± 105 mg/g at day 35. During the relocation, glycogen concentrations in foot tissue differed between locations ($p < 0.01$) and over time ($p < 0.01$), but not among treatments. Glycogen concentrations in foot tissue averaged over time and treatments were 136 ± 10 mg/g in the pond-relocated mussels and 161 ± 46 mg/g in the river-relocated mussels. Although glycogen varied between locations, concentrations in both groups were similar to those that might be expected in reference mussels. Our estimates of glycogen concentrations in *A. plicata* sampled directly from the river averaged 148 mg/g in 1996, 138 mg/g in 1997, and 116 mg/g in 1998. These results suggest that the relocation did not alter glycogen reserves in foot tissue. The most substantial change in glycogen occurred over time. In the river-relocated mussels, mean glycogen concentrations declined 44% from an average of 216 mg/g in 1995 to only 121 mg/g in 1998 (Fig. 5). In contrast, mean glycogen concentrations in the pond-relocated mussels remained similar throughout the 36 mo and ranged only from 123 mg/g to 147 mg/g.

Glycogen concentrations in mantle tissue were consistent over the quarantine period and averaged 302 ± 224 mg/g at day 0 and 277 ± 201 mg/g at day 35. During the relocation, glycogen concentrations in mantle tissue differed among combinations of location and treatment ($p < 0.01$), but not over time. Mantle glycogen concentrations were greater in mussels relocated to buried trays, suspended trays, and corrals in the Mississippi River than in those relocated similarly to the pond (Fig. 6). However, the mussels relocated to mesh bags in the pond had greater mantle glycogen concentrations than those relocated to mesh bags in the river. The lack of a temporal pattern in mantle glycogen suggests that glycogen in mantle tissue was not strongly influenced by the relocation.

The TCI of *A. plicata* differed between locations and among treatments in complicated ways (Fig. 7). The quadratic (∪-shaped) response over time differed between locations ($p < 0.01$) and the linear effect of time differed among treatments ($p < 0.01$). We included the reference mussels as a treatment in the physiological analyses, so this significant treatment × time interaction was likely driven by the slight decline in TCI in the reference mussels over time (predicted means: 3.9 in 1996, 3.8 in 1997, and 3.6 in 1998; Fig. 7). The treatment × location interaction was also significant ($p < 0.01$). The net effect of these interactions is that TCI values in mussels that were suspended from the sediments in mesh bags or trays in the pond tended to increase at a decreasing rate over time, whereas TCI values from mussels relocated into pond substrates (buried trays or corrals) tended to increase at an increasing rate (Fig. 7). Conversely, TCI values from mussels suspended over the sediments in mesh bags or trays in the river changed little over time, whereas values for mussels relocated into river substrates (buried trays or corrals) tended to decline. Last, the shape of the TCI curve over time varied be-

←

Product-limit survival estimates obtained from the fitted Cox proportional hazards model are marked at the midpoints of years and at the beginning and end of the experiment; transitions marked at September correspond to times when particular mussels that were lost to the study were last observed alive; see Methods for explanation.

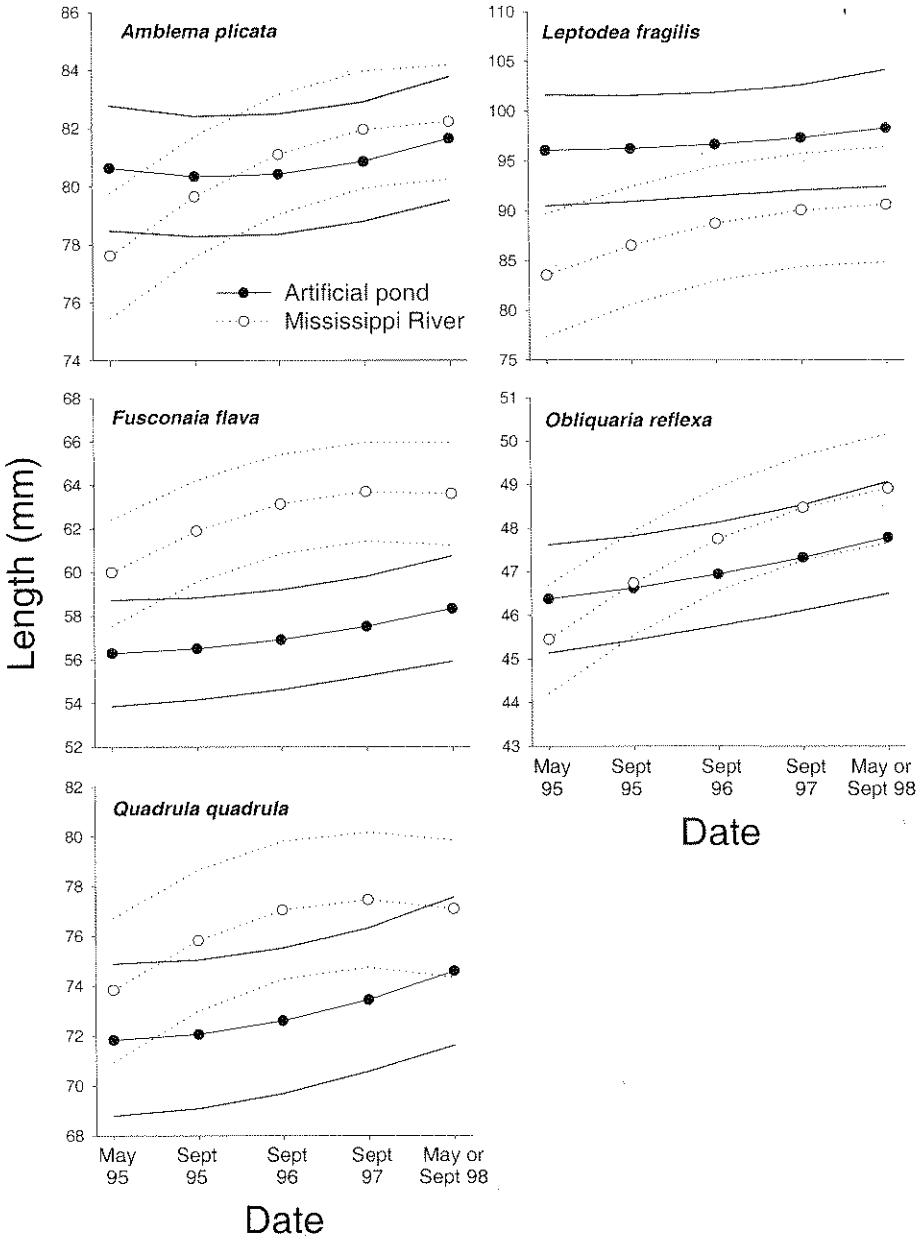

FIG. 3. Mean shell length in 5 mussel species (averaged across treatments) during a 36-mo relocation into an artificial pond or a 40-mo relocation into the Upper Mississippi River. Model-estimated values (circles) and the upper and lower 95% confidence limits (lines) are plotted for each species and location. Mussels relocated into the pond were removed in May 1998, whereas those in the river were removed in September 1998. The model-based estimates are the population-averaged best linear unbiased predictions from the individual growth trajectories; see Methods for explanation.

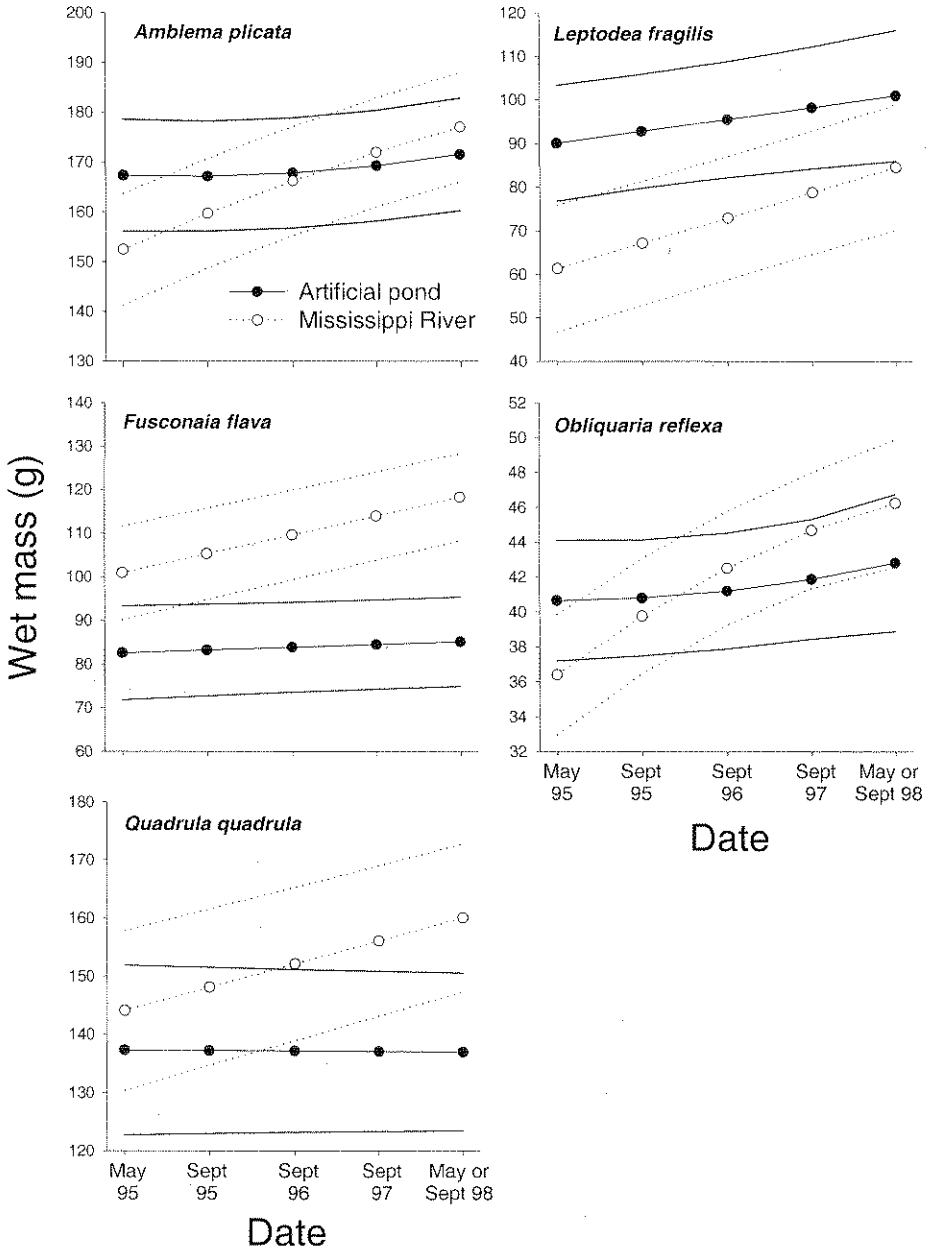

FIG. 4. Mean wet mass in 5 mussel species (averaged across treatments) during a 36-mo relocation into an artificial pond or a 40-mo relocation into the Upper Mississippi River. Model-estimated values (circles) and the upper and lower 95% confidence limits (lines) are plotted for each species and location. Mussels relocated into the pond were removed in May 1998, whereas those in the river were removed in September 1998. The model-based estimates are the population-averaged best linear unbiased predictions from the individual growth trajectories; see Methods for explanation.

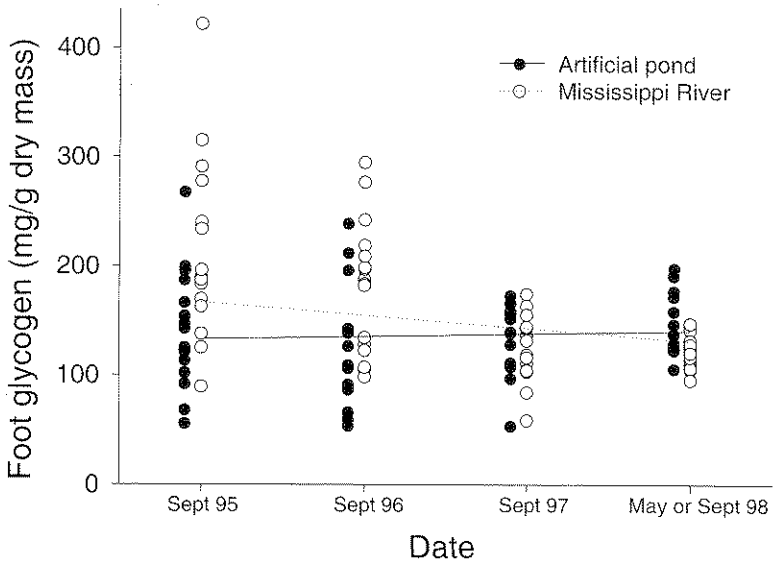

FIG. 5. Mean glycogen concentrations (circles) and predicted values (lines) in foot tissue in *Amblema plicata* (averaged across treatments) during a 36-mo relocation into an artificial pond or a 40-mo relocation into the Upper Mississippi River. Data are offset along the X-axis to better illustrate the data. Mussels relocated into the pond were removed in May 1998, whereas those in the river were removed in September 1998.

tween locations such that the relation followed a quadratic pattern in the pond-relocated mussels and was generally linear in the river-relocated mussels. However, variability was high, and these results are equivocal.

Discussion

The presence of zebra mussels in the river was a potentially confounding variable in our study. However, zebra mussel infestation levels in the river were low during this study (mean: 0.5 zebra mussel per unionid). Ricciardi et al. (1995) suggested that severe unionid mortality is not likely to occur until infestation intensity reaches 100 zebra mussels per unionid. Baker and Hornbach (1997) found that 28 zebra mussels per *A. plicata* had no effect on their condition. Haag et al. (1993) observed that 200 zebra mussels per unionid significantly reduced glycogen in *A. plicata*; these infestations were 4 times those observed in our study. Thus, it is likely that the low infestation in our study had little or no effect on the results.

A major limitation of many relocation efforts is that their success is monitored for ≤1 y (Cope and Waller 1995). In this study, 1-y survival estimates averaged 70% in the pond and 90% in the river, suggesting that short-term survival was good. Other 1-y survival estimates of pond-relocated mussels ranged from 28% (Dunn and Layzer 1997) to 84% (Gatenby 2000). However, 36 to 40 mo after relocation, survival estimates in our study were reduced to 35% in the pond and 75% in the river. These data show that 1-y survival estimates can substantially underestimate mortality observed during relocation events of longer duration.

The low survival rate of mussels relocated to the pond relative to the river was not unexpected. The species we relocated are typically found in flowing waters. Although dissolved oxygen was generally >7 mg/L near the sediment–water interface in the pond, the well water supplied few nutrients. Chlorophyll *a* concentrations in the pond averaged only 7% of those in the river. The lack of adequate food resources may have contributed to the low survival of mussels. Warmwater hatcheries in many parts of the US use local rivers as water sources; however, with the expanding distribution of the zebra mussel, many of these hatcheries may begin to experience problems with zebra mussels in their water supply. Unfortunately, this problem may preclude the use of many hatchery ponds as refugia for unionids, unless other mecha-

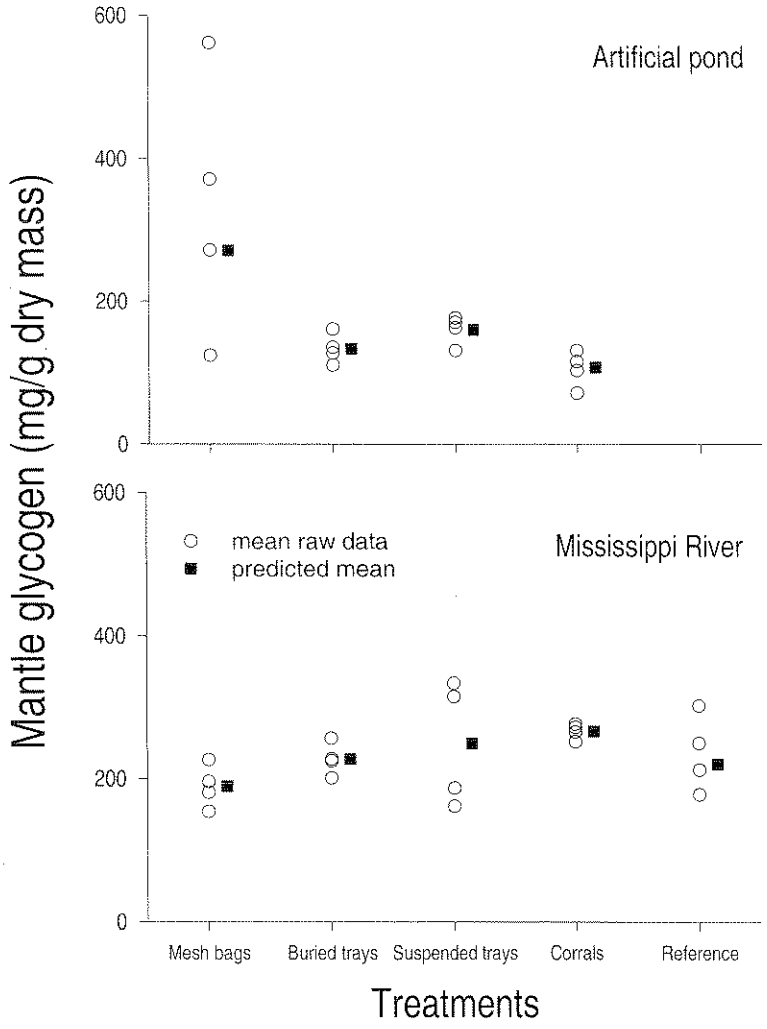

FIG. 6. Mean glycogen concentrations (circles) and predicted means (squares) in mantle tissue in *Amblema plicata* (averaged over time) during a 36-mo relocation into an artificial pond or a 40-mo relocation into the Upper Mississippi River. Mussels were placed into 1 of 4 treatments: mesh bags, buried trays, suspended trays, and corrals. Data for the reference mussels are mussels that were sampled directly from the river (i.e., not associated with the relocation) to estimate baseline conditions.

nisms to deliver nutritionally adequate water can be found.

The manner in which mussels were held influenced survival. We hypothesized that treatments that allowed mussels to burrow into sediments would promote better long-term survival; however, our data do not support this hypothesis. We observed significant differences in survival between mussels in the suspended versus buried trays; these treatments contained the same sediment but differed only in their position in the water column. This observation suggests that factors other than sediment influenced survival. Burress (1995) also observed differences in survival between mussels that were suspended compared with those placed in a cage on a pond bottom. Although the importance of sediment to mussels in providing stability, protection from predators, and as food for pedal-feeding juveniles seems intuitive, the role

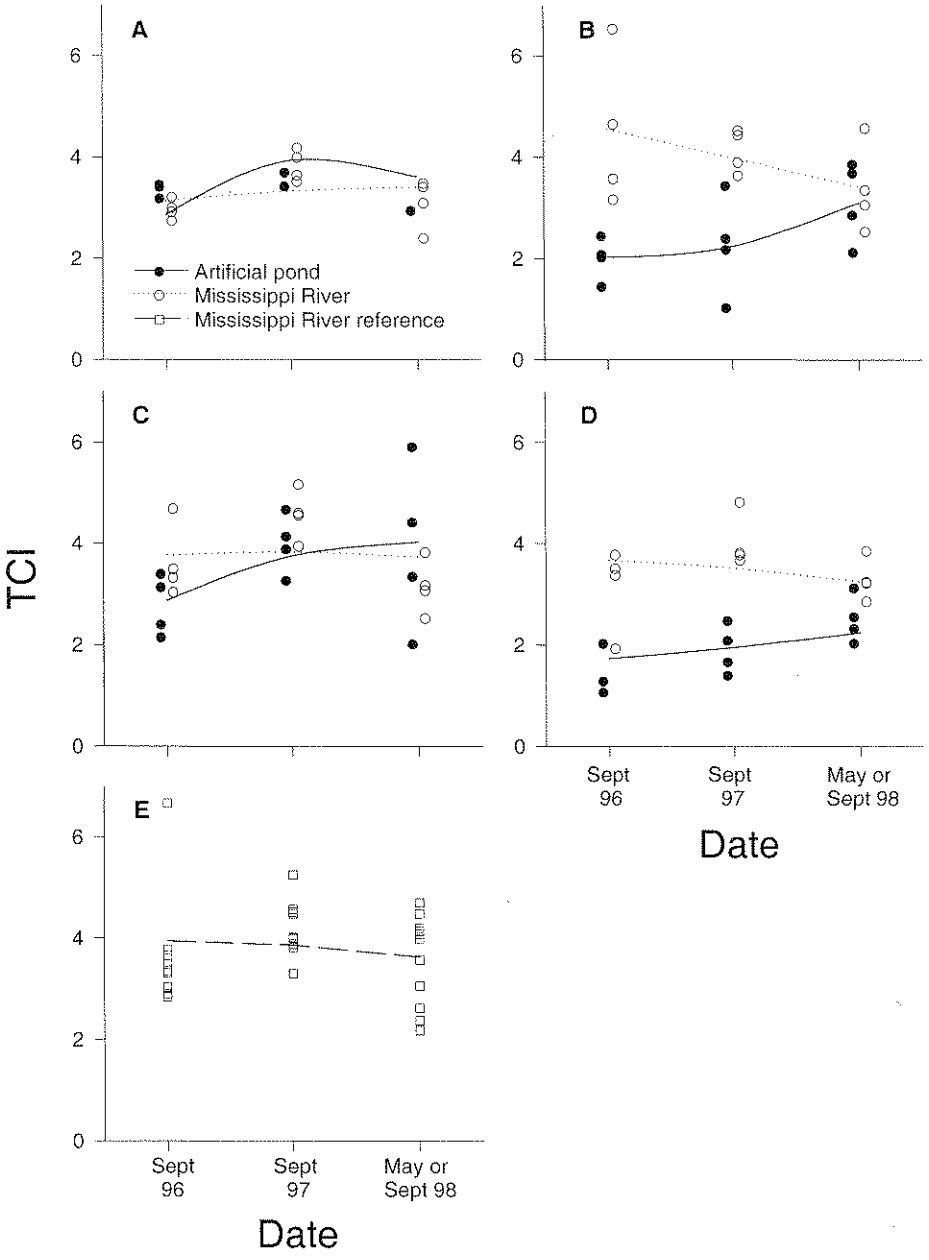

FIG. 7. Mean tissue condition index (TCI) (symbols; [tissue dry mass/shell dry mass] × 100) and predicted values (lines) in *Amblema plicata* during a 36-mo relocation into an artificial pond or a 40-mo relocation into the Upper Mississippi River. Mussels were placed into 1 of 4 treatments: (A) mesh bags, (B) buried trays, (C) suspended trays, and (D) corrals. Data for the Mississippi River reference (E) are mussels that were sampled directly from the river (i.e., not associated with the relocation) to estimate baseline conditions. Mussels relocated into the pond were removed in May 1998, whereas those in the river were removed in September 1998.

of this variable in contributing to their distribution and survival is still unclear (Holland-Bartels 1990, Strayer and Ralley 1993).

The 5 species examined in this study belong to 2 subfamilies (Ambleminae and Lampsilinae) as traditionally defined, although the phylogenetic validity of these subfamilies is still in question (Lydeard et al. 1996). Substantial differences in the morphology and physiology of mussels in these subfamilies may have contributed to the lower mortality rates in amblemines relative to lampsilines. Amblemines typically have a heavy, thick shell, whereas lampsilines generally have a thinner shell; the shells of certain lampsilines are easily crushed during handling and transportation. Many lampsiline females invest considerably more time and energy than amblemines into reproduction, and this may ultimately be manifested in differential survival rates. Other studies have documented the sensitivity of lampsilines, relative to amblemines, when examining behavioral responses to disturbance (Waller et al. 1999) and physiological responses to zebra mussels (Haag et al. 1993, Baker and Hornbach 1997).

Although we were able to document statistically significant changes in shell length and wet mass in all species over time and between locations, the magnitude of these changes, relative to the life spans of mussels, makes the utility of these measures questionable. Amblemines generally exhibit slower growth rates than lampsilines. The fastest growing species in our study (*L. fragilis*) added only 0.7 mm (pond) to 2.1 mm (river) of new shell growth each year. Changes of this magnitude are well within the ranges that have been attributed to measurement error (±1 mm; Downing and Downing 1993). Similarly, the change in mass was minimal (range: 0–2 g/y). Thus, traditional measures of growth may be inadequate endpoints in short-term relocation studies with long-lived animals.

Measures of physiological condition suggested that *A. plicata* in the pond were in poor condition compared to those relocated into the river. However, the magnitude of these differences was small relative to the variability in these measures in reference *A. plicata* in the river. In addition, no single treatment promoted better condition than the others because treatment effects were often confounded by location. Suspended mussels were generally in better physiological condition than mussels in the benthic treatments. Haag et al. (1993) showed that changes in the physiological condition of mussels may be observed before effects are manifested in mortality, but our data do not support this finding. Approximately 50% of the *A. plicata* population in the pond had died by 24 mo, yet estimates of physiological condition before these mortality events did not suggest any reduction in physiological condition. Thus, we were unable to predict mortality based on the physiological measures we chose in this species. These data suggest that although physiological measures are intuitively appealing as endpoints in relocation studies, a critical assessment of their utility is needed.

The high mortality rates, reduced growth rates, and poor physiological condition of relocated mussels in our study suggest that relocation of mussels into artificial ponds is a high-risk conservation tool. Furthermore, most midwestern mussel populations are perceived to be in immediate danger from the zebra mussel, so many relocations are being conducted quickly, with little opportunity to analyze habitat characteristics (water and sediment quality) at the destination site. There are few alternatives in certain locations and with some species; in these cases, relocation may be justified. However, based on our results and others (i.e., Cope and Waller 1995, Gatenby 2000), we strongly suggest that alternatives be explored. For example, relocations of *Margaritifera margaritifera* in Finland resulted in <50% survival after 16 y (Valovirta 1998), so it was suggested that increasing the density of parasitizing glochidia via introduction of infected fish was a more effective conservation strategy than relocation for this species (Bauer 1991). Other alternatives such as identification of fish hosts, identification of mussel beds at greatest risk from the zebra mussel, and a critical, large-scale assessment of the factors contributing to their decline may help conserve unionid populations. Clearly, more research into these areas is needed prior to the large-scale use of relocation as a conservation tool.

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Literature Cited

AMERICAN PUBLIC HEALTH ASSOCIATION (APHA), AMERICAN WATER WORKS ASSOCIATION, AND WATER ENVIRONMENT FEDERATION. 1995. Standard methods for the examination of water and wastewater. 19th edition. American Public Health Association, Washington, DC.

BAKER, S. M., AND D. J. HORNBACH. 1997. Acute physiological effects of zebra mussel (*Dreissena polymorpha*) infestation on two unionid mussels, *Actinonaias ligamentina* and *Amblema plicata*. Canadian Journal of Fisheries and Aquatic Sciences 54: 512–519.

BAUER, G. 1991. Plasticity in life history traits of the freshwater pearl mussel—consequences for the danger of extinction and for conservation measures. Pages 103–120 *in* A. Seitz and V. Loeschcke (editors). Species conservation: a population-biological approach. Birkhauser Verlag, Basel.

BURRESS, J. W. 1995. Survival and condition of riverine freshwater mussels (Unionidae) confined in cages suspended in ponds. MS Thesis, Virginia Polytechnic Institute and State University, Blacksburg, Virginia.

COPE, W. G., AND D. L. WALLER. 1995. Evaluation of freshwater mussel relocation as a conservation and management strategy. Regulated Rivers: Research and Management 11:147–155.

COX, D. R. 1972. Regression models and life tables. Journal of the Royal Statistical Society, Series B 34: 187–220.

DAVIDIAN, M., AND D. M. GILTINAN. 1995. Nonlinear models for repeated measurement data. Chapman and Hall, London, UK.

DE ZWAAN, A., AND D. I. ZANDEE. 1972. The utilization of glycogen and accumulation of some intermediates during anaerobiosis in *Mytilus edulis* L. Comparative Biochemistry and Physiology 43B: 47–54.

DOWNING, W. L., AND J. A. DOWNING. 1993. Molluscan shell growth and loss. Nature 362:506.

DUNN, C. S., AND J. B. LAYZER. 1997. Evaluation of various holding facilities for maintaining freshwater mussels in captivity. Pages 205–213 *in* K. S. Cummings, A. C. Buchanan, C. A. Mayer, and T. J. Naimo (editors). Conservation and management of freshwater mussels II: Initiatives for the future. Upper Mississippi River Conservation Committee, Rock Island, Illinois.

DUNN, H. L. 1993. Survival of unionids four years after relocation. Pages 93–99 *in* K. S. Cummings, A. C. Buchanan, and L. M. Koch (editors). Conservation and management of freshwater mussels. Upper Mississippi River Conservation Committee, Rock Island, Illinois.

DUNN, H. L., AND B. E. SIETMAN. 1997. Guidelines used in four geographically diverse unionid relocations. Pages 176–183 *in* K. S. Cummings, A. C. Buchanan, C. A. Mayer, and T. J. Naimo (editors). Conservation and management of freshwater mussels II: Initiatives for the future. Upper Mississippi River Conservation Committee, Rock Island, Illinois.

GATENBY, C. M. 2000. A study of holding conditions, feeding quantity, and suitable foods for the captive care of freshwater mussels. PhD Dissertation, Virginia Polytechnic Institute and State University, Blacksburg, Virginia.

GUY, H. P. 1969. Laboratory theory and methods for sediment analysis. Techniques of water resources investigations, Book 5, Chapter C1. US Geological Survey, Washington, DC.

HAAG, W. R., D. J. BERG, D. W. GARTON, AND J. L. FARRIS. 1993. Reduced survival and fitness in native bivalves in response to fouling by the introduced zebra mussel (*Dreissena polymorpha*) in western Lake Erie. Canadian Journal of Fisheries and Aquatic Sciences 50:13–19.

HEINRICHER, J. R., AND J. B. LAYZER. 1999. Reproduction by individuals of a nonreproducing population of *Megalonaias nervosa* (Mollusca: Unionidae) following translocation. American Midland Naturalist 141:140–148.

HEMELRAAD, J., D. A. HOLWERDA, H. J. HERWIG, AND D. I. ZANDEE. 1990. Effects of cadmium in freshwater clams. III. Interaction with energy metabolism in *Anodonta cygnea*. Archives of Environmental Contamination and Toxicology 19:699–703.

HERTZ-PICCIOTTO, I., AND B. ROCKHILL. 1997. Validity and efficiency of approximation methods for tied survival times in Cox regression. Biometrics 53: 1151–1156.

HOLLAND-BARTELS, L. E. 1990. Physical factors and their influence on the mussel fauna of a main channel border habitat of the upper Mississippi River. Journal of the North American Benthological Society 9:327–335.

HOSMER, D. W., AND S. LEMESHOW. 1999. Applied sur-

vival analysis: regression modeling of time to event data. John Wiley and Sons, New York.

HUMMEL, H., L. DE WOLF, W. ZURBURG, L. APON, R. H. BOGAARDS, AND M. VAN RUITENBURG. 1989. The glycogen content in stressed marine bivalves: the initial absence of a decrease. Comparative Biochemistry and Physiology 94B:729–733.

JOKELA, J. 1996. Within-season reproductive and somatic energy allocation in a freshwater clam, *Anodonta piscinalis*. Oecologia (Berlin) 105:167–174.

LAYZER, J. B., AND M. E. GORDON. 1993. Reintroduction of mussels into the Upper Duck River, Tennessee. Pages 89–92 *in* K. S. Cummings, A. C. Buchanan, and L. M. Koch (editors). Conservation and management of freshwater mussels. Upper Mississippi River Conservation Committee, Rock Island, Illinois.

LYDEARD, C., M. MULVEY, AND G. M. DAVIS. 1996. Molecular systematics and evolution of reproductive traits of North American freshwater unioniacean mussels (Mollusca: Bivalvia) as inferred from 16S rRNA gene sequences. Philosophical Transactions of the Royal Society of London, Biological Sciences 351:1593–1603.

MASTER, L. L., S. R. FLACK, AND B. A. STEIN. 1998. Rivers of life: critical watersheds for protecting freshwater biodiversity. The Nature Conservancy, Arlington, Virginia.

MCCULLAGH, P., AND J. A. NELDER. 1989. Generalized linear models. 2nd edition. Chapman and Hall, London, UK.

NAIMO, T. J., G. J. ATCHISON, AND L. E. HOLLAND-BARTELS. 1992. Sublethal effects of cadmium on physiological responses in the pocketbook mussel, *Lampsilis ventricosa*. Environmental Toxicology and Chemistry 11:1013–1021.

NAIMO, T. J., E. D. DAMSCHEN, R. G. RADA, AND E. M. MONROE. 1998. Nonlethal evaluation of the physiological health of unionid mussels: methods for biopsy and glycogen analysis. Journal of the North American Benthological Society 17:121–128.

NAIMO, T. J., AND E. M. MONROE. 1999. Variation in glycogen concentrations within mantle and foot tissue in *Amblema plicata plicata*: implications for tissue biopsy sampling. American Malacological Bulletin 15:51–56.

OBERMEYER, B. K. 1997. An evaluation of the Neosho River, Kansas, mussel refuge. Journal of Freshwater Ecology 12:445–452.

PLUMB, R. H. 1981. Procedures for handling and chemical analysis of sediment and water samples. Technical Report EPA/CE-81–1. Technical Committee on Criteria for Dredged and Fill Material, US Environmental Protection Agency and US Army Corps of Engineers, Vicksburg, Mississippi.

RICCIARDI, A., F. G. WHORISKEY, AND J. B. RASMUSSEN. 1995. Predicting the intensity and impact of *Dreissena* infestation on native unionid bivalves from *Dreissena* field density. Canadian Journal of Fisheries and Aquatic Sciences 52:1449–1461.

SHEEHAN, R. J., R. J. NEVES, AND H. E. KITCHEL. 1989. Fate of freshwater mussels transplanted to formerly polluted reaches of the Clinch and North Fork Holston Rivers, Virginia. Journal of Freshwater Ecology 5:139–149.

SICKEL, J. B., J. J. HEROD, AND H. N. BLALOCK. 1997. Potential for the Kentucky Dam tailwater of the Tennessee River to serve as a mussel refuge from invading zebra mussels. Pages 214–219 *in* K. S. Cummings, A. C. Buchanan, C. A. Mayer, and T. J. Naimo (editors). Conservation and management of freshwater mussels II: Initiatives for the future. Upper Mississippi River Conservation Committee, Rock Island, Illinois.

STRAYER, D. L., AND J. RALLEY. 1993. Microhabitat use by an assemblage of stream-dwelling unionaceans (Bivalvia), including two rare species of *Alasmidonta*. Journal of the North American Benthological Society 12:247–258.

TRDAN, R. J., AND W. R. HOEH. 1993. Relocation of two state-listed freshwater mussel species (*Epioblasma torulosa rangiana* and *Epioblasma triquetra*) in Michigan. Pages 100–105 *in* K. S. Cummings, A. C. Buchanan, and L. M. Koch (editors). Conservation and management of freshwater mussels. Upper Mississippi River Conservation Committee, Rock Island, Illinois.

VALOVIRTA, I. 1998. Conservation methods for populations of *Margaritifera margaritifera* (L.) in Finland. Journal of Conchology, Special Publication No. 2:251–256.

VILLELLA, R. F., T. L. KING, AND C. E. STARLIPPER. 1998. Ecological and evolutionary concerns in freshwater bivalve relocation programs. Journal of Shellfish Research 17:1407–1413.

WALLER, D. L., S. GUTREUTER, AND J. J. RACH. 1999. Behavioral responses to disturbance in freshwater mussels with implications for conservation and management. Journal of the North American Benthological Society 18:381–390.

WATTERS, G. T. 1994. An annotated bibliography of the reproduction and propagation of the Unionoidea (primarily of North America). Miscellaneous Contributions No. 1. Ohio Biological Survey, The Ohio State University, Columbus, Ohio.

WILLIAMS, J. D., M. L. WARREN, K. S. CUMMINGS, J. L. HARRIS, AND R. J. NEVES. 1993. Conservation status of freshwater mussels of the United States and Canada. Fisheries 18(9):6–22.

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