

**An Evaluation of Relocation of Unionid Mussels
Into An Artificial Pond**

Interim Progress Report for April 1, 1995 - June 30, 1995

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Submitted to:

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INTRODUCTION

The introduction of the exotic zebra mussel, *Dreissena polymorpha*, into the Great lakes and its subsequent spread into many of the nations inland river systems has been the cause of concern for public and private entities which deal with native unionid mussels. The proliferation of the zebra mussel and the documented adverse impacts they have had on native mussels prompted biologists working with the Upper Mississippi River Conservation Committee to undertake an experimental relocation project in order to identify potential conservation strategies which could be employed if zebra mussel densities in the upper Mississippi River (UMR) exhibit the same geometric increases as seen in the Illinois and Ohio rivers.

The purpose of this study was to obtain quantitative data on the growth and survival of freshwater mussels following removal from the UMR and subsequent relocation into an artificial pond. Data on growth and survival of mussels relocated to the holding pond will be compared to similar data obtained from mussels treated similarly but relocated to a natural setting in the UMR.

OBJECTIVES

This study will provide information useful for addressing the following objectives relevant to holding freshwater mussels in artificial ponds:

1. To determine if there is a difference in the growth and survival of freshwater mussels that have been relocated to an artificial pond compared to mussels held under similar conditions in the upper Mississippi River.
2. To assess species-specific differences in the growth and survival of mussels in the artificial pond.
3. To determine the mortality rate associated with removing freshwater mussels from their natural environment and subsequent placement into an artificial pond, and to determine if this rate is species-specific.
4. Once relocated, to assess which treatment (placement option of mussels into the pond or river) yields the highest growth and survival.

EXPERIMENTAL APPROACH AND DESIGN

This project consists of the removal of mussels from the UMR for a 35-day quarantine and subsequent relocation either into an artificial pond at a National Fish Hatchery or replacement back into the UMR. In either physical location, live mussels were relocated into one of four placement options (treatments). Each of these treatments were replicated four times for a total of 16 treatments per physical location. The four treatments included:

1. **Suspended substrate-filled trays.** One square meter metal baskets in a metal framework and placed about 18 to 24 inches above the substrate. These baskets were filled with dredge spoil material from the UMR that was deposited before the initial appearance of zebra mussels.
2. **Buried substrate-filled trays.** Similar to treatment 1 except these structures were placed directly on the substrate. These baskets were also filled with dredge spoil material from the UMR that was deposited before the initial appearance of zebra mussels.
3. **Corrals.** One square meter metal framework barrier around the natural substrate present in either Pool 9 of the UMR or the Genoa pond.
4. **Shoe-bag structures.** Nylon mesh bags containing 24 pockets and measuring about 20 inches by 25 inches. The bottom row of these vertically-suspended nylon structures are about 1 to 2 inches from the substrate.

All dredge spoil sediments used in treatments one and two, as well as naturally occurring sediments in the river and pond will be characterized for organic carbon and grain size.

About one half of the mussels surviving the 35 day quarantine period were relocated to an artificial pond at a National Fish Hatchery in Genoa, Wisconsin. This 0.25 acre earthen pond measures roughly 110 feet by 80 feet and is about 4 feet deep. The other half of the mussels were relocated back into Pool 9 of the Mississippi River in an area proximal to the area from which the mussels were originally obtained (River Mile 672.8 LDB).

Once relocated, all mussels will be examined twice a year (May and September) for a minimum of two years. At the time of examination, we will record survival, shell length, shell height, and wet weight for all mussels. Any dead mussels will be removed.

MUSSEL COLLECTION EFFORT

A total of 1737 mussels of seven different species were gathered from Pool 9 of the upper Mississippi River during diving operations conducted May 1-3 and May 18, 1995. During the collection efforts, unionids were thoroughly inspected and scrubbed "free" of zebra mussels. All zebra mussels attached to unionids were enumerated. The average number of zebra mussels found attached to unionid mussels was 0.7 and ranged from 0 to 24 (Table 1).

All mussels obtained during the May 1-3 collection were transported from the collection site to the quarantine pond in coolers lined with ice and layered with well-water soaked burlap bags. The quarantine pond was an 0.1 acre concrete pond at the Upper Mississippi Science Center (UMSC) that contained no inflow or outflow. Dissolved oxygen levels in the quarantine pond were maintained above 6 mg/l with an electric aerator. All unionids were hand-placed into eight net bags (about 4 ft³) that had been previously established at the UMSC (see chronological outline for details). Mortality was recorded three times a week during the 35-day quarantine period.

During the 35 day quarantine period, 387 mussels died. Survival exceeded 80% for five of the seven species (Table 2). *Truncilla truncata* (deertoe) and *Leptodea fragilis* (fragile papershell) had the lowest survival rates of 35% and 48% respectively. Overall survival of all seven species was 77%.

WATER QUALITY MONITORING

The quality of the water was measured three times a week in the quarantine pond at the UMSC, weekly in the Genoa pond, and biweekly in Pool 9 of the UMR. Monitoring of key water quality parameters may forestall potential problems before any develop. During the quarantine period, water temperature averaged 17.2°C, mean dissolved oxygen concentration was 10.9 mg/L and the pH averaged 9.0 (Table 3). Temperature and alkalinity were consistently greater in the Genoa pond compared to the River

location (Table 3). Dissolved oxygen, pH, conductivity, and hardness were similar between the pond and river.

In addition to the routine measurements mentioned above, we also contracted for a more detailed water analyses for the 0.25 acre Genoa pond prior to selecting this particular pond for the relocation effort. This water sample was taken from the 0.25 acre pond on March 21, 1995; these data are presented in Table 4.

MUSSEL RELOCATION EFFORT

On June 5 and 6, 1995, the 35-day quarantine period ended and mussels were prepared for relocation to the Genoa pond and to Pool 9 of the upper Mississippi River. In both physical locations, 24 mussels were randomly placed into each of the 16 treatments. Each treatment received five *Quadrula quadrula*, five *Obliquaria reflexa*, five *Fusconaia flava*, eight *Amblema plicata*, and three *Leptodea fragilis*. Mussels were hand-picked from the net bags in the quarantine pond, inspected carefully for zebra mussels, measured for shell length, height, and wet weight, and tagged with a unique identification number. We found no zebra mussels on any live unionids after the 35-day quarantine period. However, we inadvertently processed one dead *Amblema plicata* containing two live zebra mussels in its mantle cavity. Both zebra mussels (12 and 13 mm in shell length) were histologically examined and found to be very young males. Thus, the likelihood of newly settled veligers attaching to the mussels during the quarantine period was improbable.

A total of 768 mussels were relocated; 384 went to an artificial pond at the Genoa National Fish Hatchery and 384 were relocated back into Pool 9 of the UMR near Victory, Wisconsin. The mean shell length, shell height, and wet weight of mussels relocated into the pond and river are shown in Table 5. In general, the sizes of mussels used were similar between the pond- and river-relocated mussels. Furthermore, statistical analyses revealed there were no significant differences in mean length, height, or wet weight of the mussels that were placed into any of the treatments (i.e., the mean size of *Leptodea fragilis* placed into a "shoe bag" in Genoa was similar to the mean size of the same species placed into either a buried substrate-filled tray, suspended substrate-filled tray, or corral in Genoa; Table 6).

ACKNOWLEDGEMENTS

This progress obtained on this project has been largely a function of the generous support and hard work by many individuals representing several natural resource agencies. Specifically, a large debt of gratitude is owed to the Fisheries program of the Mississippi River Work Unit of the Wisconsin Department of Natural Resources for the many hours of underwater services they provided placing structures and collecting mussels.

This effort has been truly a cooperative venture with participation and support from ten different agencies and institutions including the staff from the Wisconsin, Iowa, Minnesota, and Missouri natural resource agencies, the U.S. Fish and Wildlife Service, the National Biological Service, Upper Mississippi Science Center, the Environmental Management Technical Center, and the Illinois Natural History Survey.

This project has received broad regional media coverage, with print, radio, and television exposure. Local television stations (WKBT) and larger market news services such as The Wisconsin State Journal, the Milwaukee Journal Sentinel, and the Minneapolis Star Tribune conducted numerous interviews with scientists working on this project (see enclosed newspaper articles).

FINANCIAL SUMMARY

The summary of our financial account between April 1, 1995 and June 30, 1995 is presented in Table 7.

Chronological List of Events for Mussel Relocation Project

- 2/22/95 Conducted the first reconnaissance dive. We identified a diverse mussel bed containing adequate densities and species diversity for this project. In addition, we obtained 60 mussels to be analyzed for viral and bacterial diseases
- 3/13/95 Laboratory assays on mussel samples identified no certifiable bacterial isolates. No viral agents incubated after 14 days.
- 3/22/95 Obtained one composite water sample from the 0.25 acre Genoa pond. This sample was sent to a contract lab for analysis.
- 4/12/95 Established temporary holding structures (net bags) at Upper Mississippi Science Center for the 35 day quarantine.
- 4/13/95 Quarantine pond filled with LaCrosse well water.
- 5/1/95- Collection of unionid mussels from Pool 9 of the UMR near Victory,
5/3/95 Wisconsin. All mussels were transferred to the quarantine pond.
- 5/18/95 Additional mussels gathered from Pool 9 of the UMR near Victory, Wisconsin and transported to the quarantine pond.
- 5/3/95- Quarantine period for all unionid mussels.
6/5/95
- 5/30/95 All metal holding structures in place in Pool 9 of the UMR.
- 5/31/95 All metal holding structures in place at Genoa pond.
- 6/5/95 Three hundred and eighty four mussels transferred from quarantine pond to the pond at Genoa, WI.
- 6/6/95 Three hundred and eighty four mussels transferred from quarantine pond to the river near Victory, WI.
- 6/20/95 Chicken wire put around all mussel-holding structures in Genoa pond to prevent mussel loss due to predation.

Table 1. Sample size, mean number, range, and standard deviation of zebra mussels found on individual unionids sampled from Pool 9 of the upper Mississippi River near Victory, Wisconsin, during May 1-3 and May 18, 1995. Means for a given species that are not accompanied by a common letter were significantly different ($\alpha = 0.05$, Tukey's *hsd* procedure).

Species	Sample Size	Mean	Range	Standard Deviation
<i>Leptodea fragilis</i>	160	0.3 ^a	0-6	0.9
<i>Truncilla truncata</i>	293	0.6 ^e	0-6	1.0
<i>Megalonaias nervosa</i>	52	2.8 ^b	0-24	3.8
<i>Amblema plicata</i>	663	0.8 ^{c,e}	0-13	1.5
<i>Quadrula quadrula</i>	199	0.9 ^{d,c}	0-10	1.7
<i>Fusconaia flava</i>	165	0.4 ^a	0-5	0.9
<i>Obliquaria reflexa</i>	205	0.2 ^a	0-6	0.7
Total	1737	--	--	--
Overall Mean	--	0.7	0-24	1.5

Table 2. Percent survival in seven species of unionid mussels after a 35-day quarantine period at the Upper Mississippi Science Center, May 1-June 5, 1995.

Species	Common Name	Sample Size on June 5, 1995	Percent Survival
<i>Leptodea fragilis</i>	Fragile papershell	83	52
<i>Truncilla truncata</i>	Deertoe	102	35
<i>Megaloniaias nervosa</i>	Washboard	52	100
<i>Amblema plicata</i>	Threeridge	624	94
<i>Quadrula quadrula</i>	Mapleleaf	192	96
<i>Fusconaia flava</i>	Pigtoe	141	85
<i>Obliquaria reflexa</i>	Threehorn wartyback	165	80
Total	--	1359	--
Overall Mean	--	--	77

Table 3. Mean temperature, dissolved oxygen, pH, conductivity, alkalinity, and hardness in water samples obtained three times a week at the quarantine pond at the Upper Mississippi Science Center (UMSC), weekly at an artificial pond at the Genoa National Fish Hatchery, and biweekly in Pool 9 of the upper Mississippi River (UMR) near Victory, Wisconsin. Data collection period April 1, 1995 through June 22, 1995.

Location	Temperature (°C)	Dissolved Oxygen (mg/L)	pH	Conductivity (μ S/cm)	Alkalinity (mg/L as CaCO ₃)	Hardness (mg/L as CaCO ₃)
Quarantine Pond at UMSC	17.2	10.9	9.0	-- ^a	-- ^a	-- ^a
Pool 9, UMR	16.0	11.0	8.2	465	148	228
Genoa Pond	20.2	11.2	8.7	457	250	264

^a Not measured.

Table 4. Characterization of water in the 0.25 acre pond at the Genoa National Fish Hatchery. All data were obtained from a single composite sample obtained on March 21, 1995.

Parameter	Concentration
5 day Biological Oxygen Demand	3 mg/L
Chemical Oxygen Demand	<6.2 mg/L
Chloride as Cl	3.6 mg/L
Nitrate as NO ₃	<0.02 mg/L
Ammonia Nitrogen	<0.07 mg/L
Total Kjeldahl Nitrogen	0.18 mg/L
Total Phosphorus	0.13 mg/L
Dissolved Phosphorus	0.12 mg/L
Total Solids	286 mg/L
Suspended Solids	<4 mg/L
Total Volatile Solids	94 mg/L
Sulfate as SO ₄	12.0 mg/L
Total Calcium	38.4 mg/L
Total Iron	0.347 mg/L
Total Magnesium	43.8 mg/L
Total Manganese	<0.006 mg/L
Total Potassium	1 mg/L
Total Sodium	2.79 mg/L
Chlorophyll <i>a</i>	4.3 μg/L

Table 5. Mean shell length, shell height, and wet weight of five species of unionid mussels relocated either to the Genoa National Fish Hatchery or to Pool 9, Upper Mississippi River. There were no significant differences among treatments in mean length, mean height, or mean wet weight within a given mussel species and location ($\alpha = 0.05$, Tukey's *h*_{sd} procedure).

Species	Treatments											
	Shoe Bags			Buried Substrate-Filled Trays			Suspended Substrate-Filled Trays			Corrals		
	Length (mm)	Height (mm)	Wet Weight (g)	Length (mm)	Height (mm)	Wet Weight (g)	Length (mm)	Height (mm)	Wet Weight (g)	Length (mm)	Height (mm)	Wet Weight (g)
<i>Mussels Relocated to Genoa National Fish Hatchery</i>												
<i>Amblema plicata</i>	79.9	61.1	163	79.7	59.6	164	83.0	61.8	177	79.3	60.1	164
<i>Fusconaia flava</i>	57.2	52.7	85	55.4	50.8	80	58.0	53.7	88	55.0	52.0	78
<i>Leptodea fragilis</i>	97.2	55.0	93	105.1	57.9	109	92.8	50.0	77	90.0	50.8	77
<i>Obliquaria reflexa</i>	47.4	40.1	44	44.7	36.7	38	46.3	38.1	35	47.2	39.4	46
<i>Quadrula quadrula</i>	75.4	65.2	152	74.9	62.9	157	66.4	57.5	109	71.1	61.5	137
<i>Mussels Relocated to Pool 9, Upper Mississippi River</i>												
<i>Amblema plicata</i>	77.6	60.3	160	76.9	58.7	146	77.0	58.1	149	78.4	59.9	155
<i>Fusconaia flava</i>	57.7	53.1	91	62.3	56.4	107	57.7	53.2	87	63.0	57.8	119
<i>Leptodea fragilis</i>	77.6	44.6	46	87.1	48.2	73	79.0	43.5	52	89.9	49.3	73
<i>Obliquaria reflexa</i>	45.5	37.8	36	45.8	38.7	39	46.9	38.7	39	43.6	35.8	34
<i>Quadrula quadrula</i>	72.6	62.3	144	74.7	65.4	144	74.2	64.1	148	74.4	64.6	149

Table 6. Mean (± 1 SEM) shell length, shell height, and wet weight of five species of unionid mussels relocated either to the Genoa National Fish Hatchery or to Pool 9, Upper Mississippi River.

Species	Shell Length (mm)	Shell Height (mm)	Wet Weight (grams)
<i>Mussels Relocated to Genoa National Fish Hatchery</i>			
<i>Amblema plicata</i>	80.5 \pm 1.1	60.6 \pm 0.8	167 \pm 6
<i>Fusconaia flava</i>	56.4 \pm 1.1	52.3 \pm 1.0	83 \pm 4
<i>Leptodea fragilis</i>	96.1 \pm 2.7	53.2 \pm 1.4	89 \pm 7
<i>Obliquaria reflexa</i>	46.4 \pm 0.6	38.6 \pm 0.6	41 \pm 2
<i>Quadrula quadrula</i>	72.0 \pm 1.7	61.8 \pm 1.4	139 \pm 8
<i>Mussels Relocated to Pool 9, Upper Mississippi River</i>			
<i>Amblema plicata</i>	77.5 \pm 1.1	59.3 \pm 0.7	153 \pm 5
<i>Fusconaia flava</i>	60.2 \pm 1.3	55.1 \pm 1.2	101 \pm 6
<i>Leptodea fragilis</i>	83.7 \pm 3.1	46.5 \pm 1.9	62 \pm 7
<i>Obliquaria reflexa</i>	45.4 \pm 0.6	37.8 \pm 0.6	37 \pm 2
<i>Quadrula quadrula</i>	73.9 \pm 1.3	64.1 \pm 1.1	146 \pm 7

Table 7. Financial record of the mussel relocation project account between April 1, 1995 and June 30, 1995.

Transaction	Credit	Debit	Balance
<i>Shell Exporters of America and Mussel Mitigation Trust Account</i>			
Donations from Shell Exporters of America and Mussel Mitigation Trust	\$7061.37	--	--
Tagging and quarantine supplies	--	\$493.22	--
River and pond supplies	--	\$4401.81	--
Zebra mussel samplers	--	\$350.00	--
Water quality analysis on the 0.25 acre pond at Genoa, performed by a contract laboratory	--	\$360.00	--
Total balance in account	--	--	\$1456.34
<i>Mississippi Interstate Cooperative Resource Association Account</i>			
Donation from Mississippi Interstate Cooperative Resource Association	\$20,000	--	--
Contractual diving costs	--	\$5200.00	--
Supplies	--	\$394.00	--
Total balance in account on June 30, 1995	--	--	\$14,406.00
Total project balance on June 30, 1995	\$27,061.37	--	\$15,862.34