Lampsilis higginsii recovery project
Genoa National Fish Hatchery
2004

Roger Gordon and Tony Brady
January 10, 2004
Genoa National Fish Hatchery

Production activities carried out at Genoa National Fish Hatchery during 2004 for the federally endangered Higgins Eye Pearlymussel (*Lampsilis higginsii)* represent the fifth year of a multi-year effort to re-establish and enhance populations of this rare mussel within the Upper Mississippi River watershed. Funding for this project was provided by the U.S. Army Corps of Engineers, and the U.S. Fish and Wildlife Service, Fisheries Division. Cooperating partners included the State of Minnesota Department of Natural Resources, State of Wisconsin Department of Natural Resources, State Illinois Department of Natural Resources, and the State of Iowa Department of Natural Resources.

The goal of this project is to produce large quantities of healthy juvenile and sub adult *L. higginsii* for reintroduction into suitable habitats within its current and former range. Specific habitats for reintroduction or cage propagation included twelve areas identified by the 2004 Higgins Eye Pearlymussel Conservation Work Plan. These areas included sites within the Mississippi R. watershed in Iowa, Wisconsin, Minnesota, and Illinois (fig 1). Due to the lack of historical intensive propagation success with this species, the focus of production efforts during 2004 was extensive in nature, with the majority of juvenile mussels' excysting in natural environs. The following report is a synopsis of the major activities performed through the Genoa National Fish Hatchery during the calendar year 2004. Categories for discussion include: 1. Host fish production. 2. Infestation and incubation. 3. Host fish cage project and free releases. 4. Future program

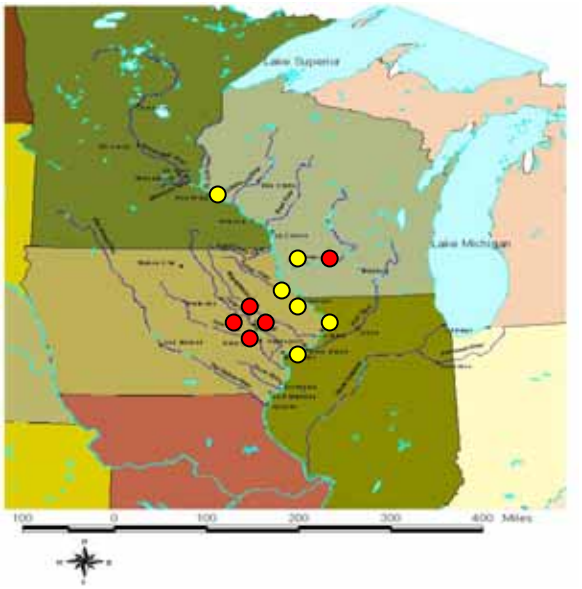


Fig. 1. *L. higginsii* cage culture (yellow), and free release (red) sites in 2004.

Host Fish Production

The production of host fish for use in the propagation of Unionids is the first step in any successful freshwater mussel culture program. Genoa National Fish Hatchery has the capability to produce many of the species of known host fishes for various mussel species found in the Upper Mississippi River watershed. The facility produced and used two known host fish species for this project during 2004. Largemouth bass (*Micropterus salmoides)* and smallmouth bass (*Micropterus dolomieui)* were selected based on juvenile mussel production histories for these fish on the facility, as well as the ability of these particular centrarchids to tolerate the stresses associated with the cage culture portion of the project. Walleye (*Sander vitreus)* provided by the Iowa Department of Natural Resources, were also infested by hatchery staff during October 2004 for free release operations carried out at Central City, IA. Numbers of fish inoculated and released during the spring 2004 infestation operation included 3828 largemouth bass and 4,430 smallmouth bass for a total of 8,258 fish.

Fish used during fall field infestations in 2004 included an additional 629 smallmouth bass, 1058 walleye, and 60 largemouth bass.

Fig. 2. *Lampsilis higginsii* infested Largemouth Bass yearling at Genoa National Fish Hatchery

Post attachment survival of host fish continues to improve over initial values for the project. Survival during 2004 was 98.7% as compared with 97.8% for 2002. Increased survival of infested fish has been directly related to a more aggressive disease control policy adopted after the 2002 production season where project losses to columnaris infections (causative agent *Flexibacter columnaris*) post infestation were as high as 20% for some lots. Joint USGS/FWS trials conducted by the Upper Midwest Environmental Science Center, LaCrosse and Genoa National Fish Hatchery during 2004 has shown no effect on survival of encysted mussel treated with several common theraputant's for controlling external columnaris infections as well as the management of external parasite infestations. Data from this study and another to be carried out during FY 2005 should provide further guidance for managing pathogens experienced during the culture period.

The project continued to recover from depopulation of all Centrarchid stocks during 2002 due to the detection of the causative agent for largemouth bass virus (LMBV) being detected in fall fingerling stocks on the hatchery, (see 2003 report). The destruction of all centrarchid broodstocks, (largemouth bass, smallmouth bass, bluegill *Lepomis macrochirus*, and black crappie *Pomoxis nigromaculatus*) during 2002 did impact production for 2003, but suitable Mississippi watershed stocks have been reconstituted and production levels during 2004 were at pre-detection levels. Genoa NFH will continue to augment brood populations through the introduction of wild fish over the next several years to ensure adequate genetic representation of donor populations in captive stock. All captive stocks will continue to be subjected to rigorous annual health inspections to reduce the possibility of the distribution of fish pathogens within the watershed by activities of this or other programs being carried out by Genoa NFH. Despite setbacks the annual production of host fish for *L. higginsii* restoration has continued to grow over the life of the project and is projected to continue to expand in FY 2005 (fig. 3).

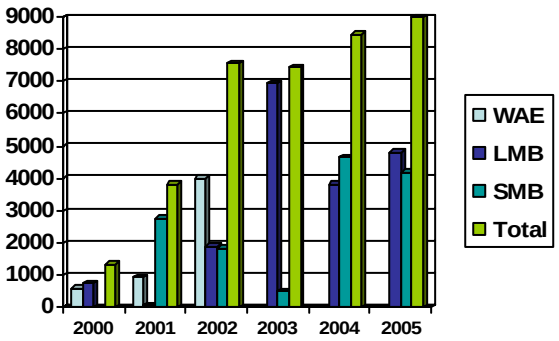


Fig.3. Annual host fish production at Genoa NFH for *L. higginsii* project

Infestation and Incubation

Mussel infestation operations during spring 2004 infested 8,470 fish as compared to 7,440 in 2003. As in 2003

largemouth bass and smallmouth bass were selected as hosts, based on past performance for survival and numbers of transformers produced. Estimated numbers of spring juveniles produced also increased for the fifth consecutive year with 545,678 juveniles' transformed (fig. 4). While total reportable numbers of juveniles produced did increase slightly during 2004, average number of mussels excysted per fish declined to 66.08.

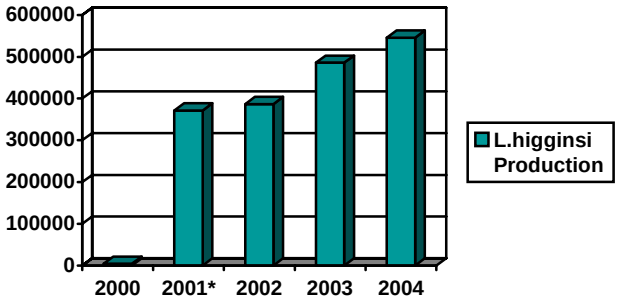


Fig. 4. Juvenile production estimates for *L. higginsii* from Genoa culture facility

Although number of excysted mussels produced per fish did decline in 2004, initial attachment rates increased over past years with average attachment over all lots at 373 mussels/fish (table 1). Past transformation data has shown that *L. higginsii* typically transform at a rate of 40-60 % for spring infestation events. Excystment percentages for *L. higginsii* were markedly lower than these historical values at ~17.7 %. An investigation of the aquaria battery used to monitor transformation timing and values revealed system modifications carried out in late 2003 allowed escapement of juveniles from the collection tanks into the filter system. Removal of entrained juveniles from the diatomaceous earth system was not possible or practical and strain integrity was not maintained. Based on escapement evidence it is certain that overall transformation rates were considerable higher than reported values, and consistent with past percentages.

Strain	# Donor ♀'s	# Fish / Species	Mussels/ Fish	Est. Juv. Produced
St. Croix	5	339 / LMB	416.4	22,400
Cordova	19	2614 / LMB	418.0	138,104
Cordova	23	2873 / SMB	285.3	224,473
Cassville	8	875 / LMB	299.2	57,818
Cassville	8	1022 /SMB	428.4	67,531
Wisconsin	3	535 / SMB	395.8	35,352
Totals	**66**	**8,258**		**545,678**

Table 1. Production values for spring 2004 *L. higginsii* project at Genoa NFH.

The total production values expressed in the above table do not include fish and mussel numbers for field operations carried out in October of 2004 in eastern Iowa. Fall field inoculations were carried out on the Wapsipinicon River near Central City IA, on October 14 in cooperation with Iowa Department of Natural Resources Fisheries division. A combination of hatchery produced and wild fishes were inoculated with the larvae of 12 Cordova strain *L. higginsii* females and released locally (table 2).

Female mussels used in fall operations were collected from an aggregation site in pool 14 of the Mississippi River by hatchery and state divers. Donor mussels were held at the Genoa NFH and transported to the inoculation site on the day of the event. After harvest all mussels were returned to the collection site and released. No mortalities were observed and all mussels were marked for future evaluation. Additionally, genetic samples in the form of mantle clips were taken from donor females for future analysis. Juvenile excystment projections for the fall operation were calculated using counts of attached glochidia collected from gill samples of infested fish (table 2). A rate of excystment was calculated using average production values of vernal infestation trials carried out during 2000-2002 for the same species of hosts.

Number Fish Infested	Source	Release Site UTM 15T	Est. Attach.	Est. Trans. 49.7%
500 SMB	Genoa NFH	0621823 4673111	101,250	50,321
129 SMB	Wild	0620405 4674751	26,122	12,982
1050 WAE	Rathbun SFH	0621823 4673111	212,625	105,674
60 LMB	Wild	0620405 4674751	12,150	6,038
8 WAE	Wild	0620405 4674751	1,620	805
Totals 1,747			**353,941**	**175,820**

Table 2. Production estimates for fall 2004 field infestations carried out at Central City, IA. Estimates based on field and lab studies carried out 2000-2002.

Donor female *L. higginsii* mussels used during 2004 operations were collected from four populations within the upper Mississippi R. watershed. Female mussels were collected from the St. Croix River (Hudson Narrows), Mississippi River populations at Cassville, WI. and Cordova, IL. and from the Wisconsin River near Prairie du Sac. Total numbers of mussels harvested was 66, a decrease from 76 individuals used in 2003 (table 1). All mussels were collected by state or federal divers and arrived at the culture facility in excellent condition. All females were gravid, and harvested glochidia appeared mature. No intermediate larvae were detected and all sampled glochidia responded to standard NaCl viability tests.

Fig. 5. Captive female *L. higginsii* displaying mantle lure held at Genoa NFH mussel propagation facility. Photo: Mike Davis, MNDNR.

Donor mussels were returned to collection sites after glochidia harvest and appeared in good condition with no mortalities or morbid individuals observed. Genetic samples consisting of mantle sections were collected from all mussels during 2004. These samples were preserved in 90% ethyl alcohol and are being stored at the Upper Midwest Environmental Science Center, Lacrosse, Wisconsin.

All fish infested during spring 2004 operations were held at the facility for a minimum of two weeks post exposure to ensure encystment of attached juveniles and reduce fish losses to stress and predation. After this incubation period those fish designated for release were removed from the holding facility and transferred by hatchery personnel to pre-determined plant sites within the Mississippi River watershed. The proportion of total fish used for river excystment constituted the majority of host fish in this project, with over 99% of surviving fish releasing their mussel compliment within the watershed. The remaining fish were held at the facility to monitor rate of transformation and provide excystment values to be applied to free release and cage programs.

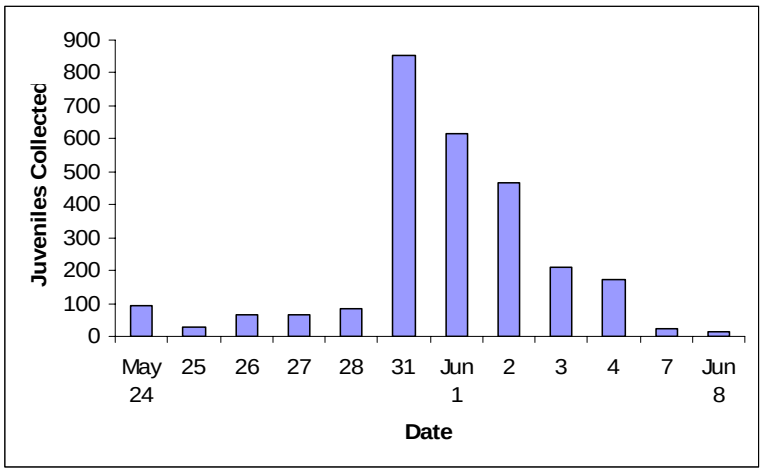


Fig. 6. Juvenile *L. higginsii* collected from monitoring aquaria at Genoa NFH mussel propagation facility.

Figure 6 describes number of juveniles excysted over time for all strains of *L. higginsii* and hosts at the Genoa Mussel Propagation Facility during spring of 2004. Values do not reflect escapement of juveniles that were later recovered.

Host Fish Caging Project

In an effort to expand on the successes of the 2003 field season cage propagation was expanded from 89 cages at 6 sites in the Mississippi R. watershed to 148 cages at 9 sites (table3). Most of the expansion was in the form of the "closed bottom" cage design first deployed in 2001 (Gordon 2001).

Fig. 7. Standard closed bottom mussel excystment cage deployed in *L .higginsii* recovery project

Crews constructed all additional cages at the Genoa NFH during winter months of 2004. Noted modifications made during 2004 included the removal of the bottom screen portion of those cages designated as "closed", (those cages possessing a catch basin). This was done to alleviate small percentages of sub-adults that were impinged between the wooden floor of the base and the wire mesh of the upper cage assembly, causing deformation of shells (fig 8).

Fig. 8. *L. higginsii* sub-adult (top) harvested in fall 2003, showing cage impingement deformations.

Location	Coordinates UTM	#Cages/Type	#/Fish	Est. Trans.	Strain
Miss. R. Frontenac	15T-0554301 4929177	10 / closed	339/LMB	22,400	St. Croix
Miss. R. Lake City	15T-0559896 4920053	48 / closed	600/LMB 875/LMB	39,647 57,818	Cordova Cassville
Miss. R. Pool 12	15T-0714402 4683753	30 / closed	890/LMB	58,810	Cordova
Miss. R. Pool 16	15T-0674867 4589011	30 /closed	524/LMB 175/SMB	46,189	Cordova
Wis. R. Orion Upper	15T-0715643 4787597	10 / open	406/SMB	26,828	Cassville
Wis. R. Orion Lower	15T-0713923 4787274	11 / open	446/SMB	29,471	Cassville
Wis. R. Orion Sta. 3	15T-0714410 4787008	6/ closed	140/SMB	9,251	WI. River
Miss. R. Guttenburg Floating	15T-0655830 4738598	1 floating	110/SMB	7,268	Cassville
Miss. R. Dubuque Floating	15T-0692155 4707486	2 floating	60/SMB	3,964	Cassville
Total		**148**	**4,565**	**301,646**	

Table 3. Cage locations, numbers and estimated transformers produced.

Results from the solid bottom cages were mixed across recovery units again during 2004. Efforts carried out in Minnesota waters of Lake Pepin in pool 4 of the Mississippi R. were highly variable between cages and strains (table 3). Cordova strain cages yielded the lowest returns of the three cohorts with only 15 age 0 sub-adults harvested from 20 cages. Cassville strain cages yielded 895 fall sub-

adults from 28 units, but like the Cordova cages, production was highly variable with recoveries ranging from 0-91 mussels per cage. Both these strains were propagated at the Lake City site, a previously untested location, therefore production values are not directly comparable to past production successes using the St. Croix strain. As in 2002 and 2003 the Frontenac site of Lake Pepin was selected for cages of the St. Croix strain. St. Croix yields for 2004 were comparable to the levels of 2003, with the 2004 harvest averaging 329 mussels/cage. Due to the small size of mussels collected in 2004 only 5 of the 10

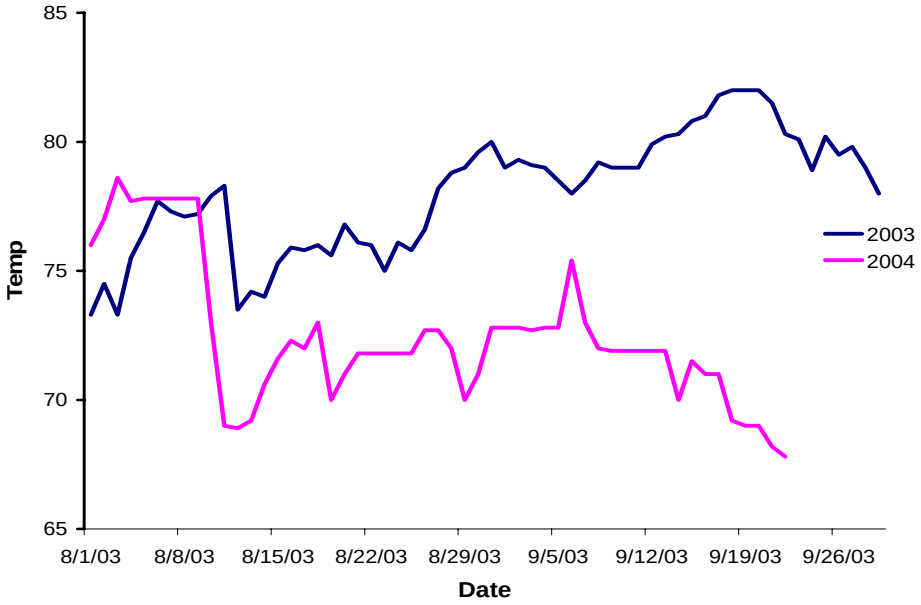


Fig. 9. Average daily water temperatures of Pool 4 of the Mississippi River for August/September 2003-2004.

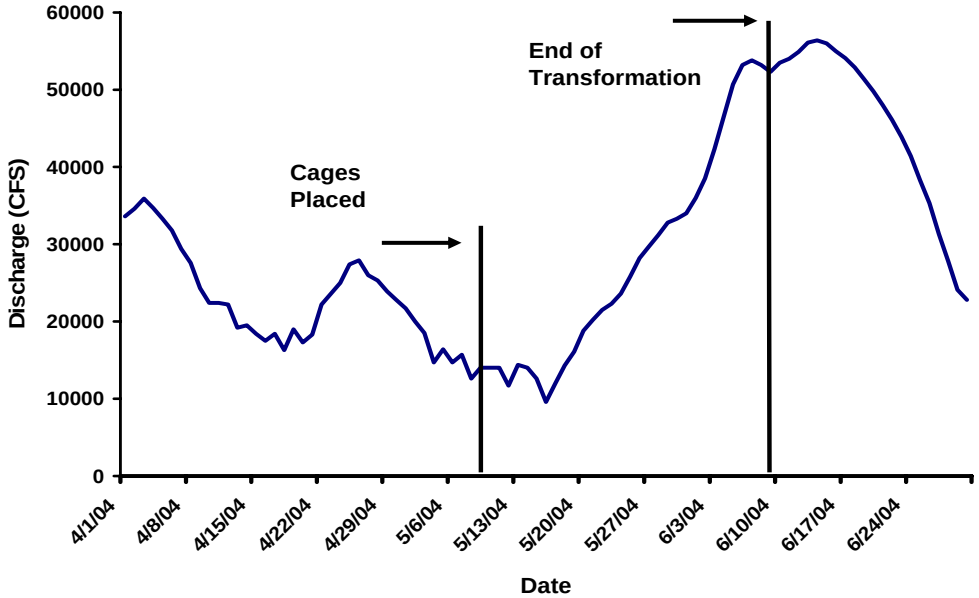


Fig. 10. Comparison of recorded discharge at Lock and Dam No. 4, Mississippi River, and projected excystment dates for L. higginsii bearing hosts fish in cages at Frontenac and Lake City, MN. 2004.

propagation cages were harvested, with the remaining 5 being deferred until summer 2005. Temperature comparisons of pool 4 discharges between 2003 and 2004 show a marked reduction of available temperature units during August/September (fig 9). These lower temperatures may have slowed growth on caged mussels during 2004. All cages at both the Lake City and Frontenac sites were moderately colonized by zebra mussels (*Dreissena polymorpha*), and the majority of the cages at the Lake City site had silt deposits within the cages of 8-10 cm. Comparing average discharge of Pool 4, (Lake Pepin) to expected date of excystment shows that the newly transformed mussels would have been exposed to a high flow event just after dropping from their host (fig 10). These high flow events carry large loads of silt and debris and may have been the source of the sediments observed in October. In addition to silt loads inherent to the main stem of the Mississippi River, the Lake City site was positioned .3 km downstream of the outflow of Miller Creek, a small tributary to the main river. Miller Creek was observed by project personnel carrying significant levels of sediment during run-off events. This combination of factors may have effected survival of excysted juveniles at the lower site.

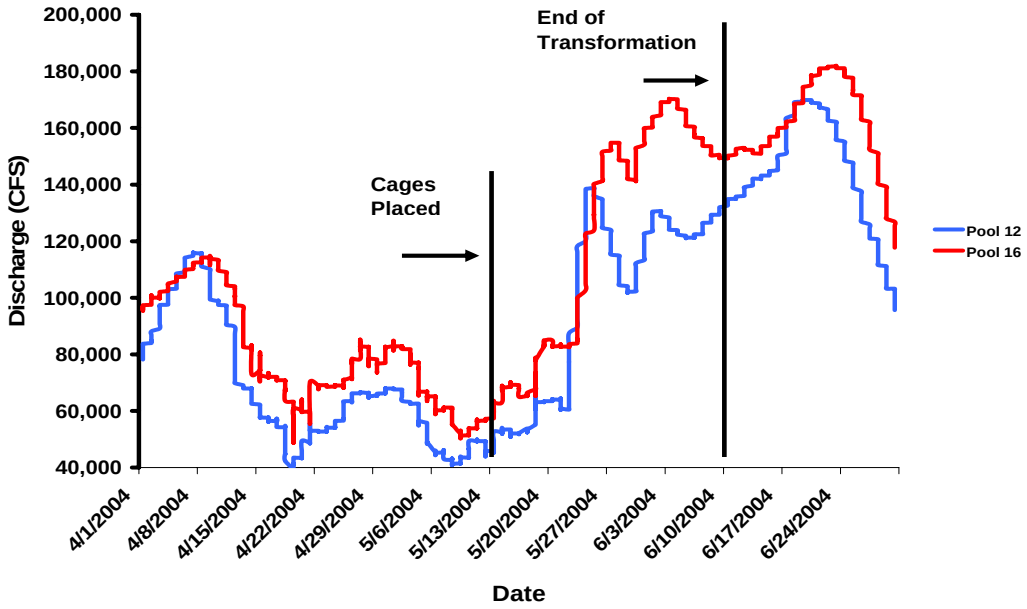
In a repeat of 2003 efforts, several additional sites outside of Lake Pepin were selected for cage propagation. Illinois DNR selected a site in pool 12, just upstream of Lock and Dam No.12 at RM 557.5. This site, located in a side slough off the main channel, was inspected by FWS divers prior to cage placement for presence of mussels and substrate stability. Divers collected over 12 species of mussels from the site, which was composed of a firm sand/silt/ cobble substrate. A second site was selected by the Iowa DNR in Pool 16 at RM 462.9. This site was also inspected by FWS divers for the presence of mussels and substrate stability. Fourteen species of mussels were collected from the immediate area which was composed of firm sand/shell/cobble. Thirty closed bottom cages were placed at both sites on May 13/14, 2004 (table 3). Results from both of these sites were unsatisfactory, with no age 0 sub-adult *L. higginsii* harvested. All cages at these sites experienced very high flows just as juveniles were expected to excyst, which may have scoured any juveniles from the cages (fig. 11). All cages had silt/debris deposits from 8-20cm deep within the catch basin as seen in figure 11.

Fig. 11. Photo of *L. higginsii* culture cage from pool 16 site showing silt deposition.

No living Unionids were found in any of the cages at the pool 16 site. The pool 12 site did yield four species of mussels, *Arcidens confragosa*, *Obliquaria reflexa*, *Utterbeckia imbecillis*, and *Truncilla donaciformis*. As in pool 16 no *L. higginsii* were collected.

Cage excystment efforts carried out on the Wisconsin River in cooperation with Wisconsin DNR were accomplished on May 11, 12, 2004.

These operations, carried out near Orion, WI. were located on the same site(s) as the efforts in 2001-2003 (table 3). Twenty-one open bottom cages were placed at the Orion site(s), with an additional 6 closed bottomed cages placed in a third site at RM 48.66. Assessment of open bottom sites 44 days post placement revealed two cages were lost. Investigation of the closed bottom site revealed that 4 of 6 closed cages were lost to high water events. Specific analysis of fish and conditions can be found in the Wisconsin DNR 2004 report to the Mussel Conservation Team (Heath 2004). Fall screening of remaining closed bottom cages yielded no *L. higginsii* sub-adults. In addition to standard excystment cages, two floating cage designs were employed at two sites in pools 11 and 12 during 2004. The pool 11 site, located on the downstream side of Lock and Dam No. 10, was the same site as used in 2003. This cage was of the same design as that used in 2003, with the addition of additional floatation. This site yielded no *L. higginsii* sub-adults when inspected on September 14, 2004. Lack of production was attributed to the cage being grounded when water levels dropped sometime after fish being introduced. The cage, located in a static cove at the base of the dam, was filled with a heavy deposit of silt up to 12 cm deep. Conditions in the cage appeared anoxic upon removal, with very low numbers of benthic invertebrates present. At a second site located at Ice Harbor, Dubuque, IA. RM 570.75, a smaller version of the pool 11 prototype floating cage was used. (fig 13). Two cages were placed in the harbor on May 15, 2004 by hatchery personnel and harvested on September 14, 2004. These cages yielded 106 age 0 sub-adult *L. higginsii* ranging in size from 2-15mm. Sub-adults produced in these cages were transferred to Lake Pepin and consolidated with other Cassville strain mussels produced at the Lake City site.



.

Fig. 12. Recorded discharges at Lock & Dam No. 12 and 16 on the Mississippi River in relation to expected *L. higginsii* excystment in culture cages.

Fig. 13. Floating mussel propagation cage located in Ice Harbor, Dubuque, IA. 2004.

Fig. 14. *L. higginsii* recovered from floating cages at Dubuque, IA. site.

Host Fish Free Release Program

Another method used during 2004 to increase the numbers of transformed juveniles introduced into the wild was the release of *L. higginsii* bearing host fish directly into areas of the watershed thought to be conducive to long term mussel survival. This practice of free release has been carried out since the early 1900's as a management tool to increase Unionid populations. The effectiveness of past operations was not readily ascertainable due to large existing native mussel populations' endemic to release waters. *L. higginsii* populations in areas used for releases during 2001-2004 were historically at detectable levels but at present are very low or absent from recent surveys. Population increases in the immediate future for this species in areas within or adjacent to release sites may be attributable to the current reintroduction program. Table 4 below describes numbers of fish and areas stocked with glochidial bearing fish and the projected resulting juvenile mussels released for spring 2004.

Release Site	Location UTM	# /Species	Est. Trans. Juveniles
WI. R. Prairie du Sac	16T-0279102 4794164	395/SMB	26,101
Wapsi. R. IA. Anamosa	15T-0641600 4662087	684/SMB	45,198
Wapsi. R. IA. Central City	15T-0621823 4673111	684/SMB	45,198
Cedar R. IA.	15T-0624480 4639887	1330/SMB	87,888
Iowa City IA.	15T-0621987 4614786	600/LMB	39,647
Totals		3529	244,032

Table 4. Free release site locations, numbers, and estimated transformed juveniles produced spring, 2004.

Diving Support Activities

Genoa NFH staff divers participated in a wide range of diving activities in support of project goals. Staff divers carried out donor female collections, cage site assessments, and culture cage placement and retrieval. Hatchery divers also assisted state cooperators in *L. higginsii* sub-adult stocking operations and adult assessment and cleaning/aggregation activities.

Future Plans

Station plans call for continued support of *L. higginsii* restoration efforts through 2005. In line with methods and management goals established in 2001, more than 98% of infested fish will continue to be used in cage culture and free release strategies for grow out of juvenile mussels. Hatchery personnel will carry out continued

excystment trials during spring 2005 to better increase understandings of developmental temperature requirements for *L. higginsii*. Additionally, the floating cage culture portion of the project will be expanded in the hopes that grow-out habitats in the lower pools within the species range may be used for production. Submerged cage culture operations in the lower pools, which has not shown promise over the past two seasons will be shifted north into pool 4, where success has occurred over the past two seasons. Genoa biologists will conduct a marking efficacy study in summer of 2005 to test the prospects of chemically mass marking *L. higginsii* sub-adults for future relocation efforts. The fatmucket mussel (*Lampsilis siliquoidea*) will be used as a surrogate for this marking trial.

Fig. 15. Progeny of 2002 cage culture operations being measured prior to release into Mississippi River in fall 2004.

Acknowledgements
Special thanks to all personnel associated with the successful propagation season of 2004. Special thanks to Illinois DNR biologists Dan Sallee, Iowa DNR biologist Scott Gritters, Wisconsin DNR biologist Dave Heath, U.S. Army Corp of Engineers biologists Dennis Anderson and Dan Kelner and Minnesota DNR biologist Mike Davis for the exceptional cooperation and assistance given to Genoa personnel throughout the past field season.

Resources

Davis, M. 2003. Clam Chronicles – 2003. Minnesota Department of Natural Resources. Lake City, Minnesota. 35pp.

Gordon, R. 2001. Operational Procedures and Techniques for the Artificial Infestation of *Lampsilis higginsii*. U.S. Fish and Wildlife Service, Genoa National Fish Hatchery, Genoa, Wisconsin. 9 pp.

Gordon, R. and T. Brady. 2003. *Lampsilis higginsii* recovery project. U.S. Fish and Wildlife Service, Genoa National Fish Hatchery, Genoa, Wisconsin. 10pp.

Heath, D.J. 2004. Report on 2004 Placement of Cages Containing *Lampsilis higginsii* in the Wisconsin River near Orion. Wisconsin Department of Natural Resources, LaCrosse, Wisconsin. 4pp.