Progress Report for USFWS Region 3, USGS Science Support Partnership Proposal

Fish Host Identification for Glochidia of the Endangered Winged Mapleleaf Mussel *(Quadrula fragosa)* at the Upper Midwest Environmental Sciences Center

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Problem Statement

The winged mapleleaf mussel (*Quadrula fragosa*), a federally listed endangered species since 1991, exists only as a remnant population in a 20-kilometer reach of the lower St. Croix River bordering Minnesota and Wisconsin. Extensive surveys of this riverine reach between 1988 and 1992 found only 77 individuals. Recruitment to this population has been very low in recent years and is dominated by the 1984 year class. One of the primary factors identified by the Winged Mapleleaf Recovery Team as limiting this population is a lack of knowledge of the mussel's life history. For example, as recently as 1997, this species' brooding period was unknown and presumed to extend from late May to mid July. Since then however, repeated observations of individuals in the St. Croix River have revealed that their brooding period more likely occurs from mid September to early October, which is quite different from that of other species in the same genera.

Another critical piece of life history information that remains unknown, and is one of the most serious obstacles in winged mapleleaf recovery efforts, is the identification of the host fish species needed for this mussel to complete its life cycle. Since 2000, investigators at the University of Minnesota's USGS Cooperative Fish and Wildlife Research Unit have received Science Support funding to determine the host fish(es) for the winged mapleleaf using species that historically occurred in the St. Croix River. Laboratory tests conducted here to date have not identified suitable host fish species for the winged mapleleaf and are continuing. However, the host fish testing program at the University of Minnesota is constrained by the limited quantity and quality of its water supply and by limited laboratory space. Combinations of these factors have reduced the number of fish species that can be tested, jeopardized the health of some fish, and limited the range of water temperatures that can be tested. As a consequence, additional tests are often necessary to demonstrate with greater certainty

whether a particular fish species is a suitable host for winged mapleleaf glochidia. Continued annual delays in the identification of the winged mapleleaf's host fish(es) represent a serious impediment in the recovery plan for this endangered species and fail to reduce the risk of extinction for its only remaining population.

Progress and Results in 2002

The winged mapleleaf interagency dive team located and recovered only one female in the St. Croix River (#390) that exhibited any signs of gravidity during searches over a 3-week period in the fall of 2002. No more than about 300 apparently mature glochidia were recovered from this individual by Mark Hove at the University of Minnesota on the evening of October 3, 2002. On October 4, about 150 glochidia were transported in tempered (16-19°C), aerated water to the Upper Midwest Environmental Sciences Center in La Crosse, Wisconsin, for fish host testing. Upon arrival, a subsample of the glochidia (about 30 individuals) were transferred to a petri dish and sodium chloride crystals were added to test for glochidial viability. All the glochidia exposed in this manner quickly responded by snapping their valves shut, indicating they were suitable for use in the planned fish host tests. The remaining glochidia were randomly sorted into five groups with about 30 individuals each.

Although five lots of fish were available for testing (flathead catfish, blue catfish, slender madtom, small channel catfish, and large channel catfish), a decision was made to only use the large channel catfish because of the small number of glochidia that were available, and the earlier success by others who successfully transformed a small number of winged mapleleaf glochidia on this species. Thus, about 30 glochidia were pipetted onto the left and right gills of each of five large channel catfish. Water from the inoculation procedure was recovered, placed in a 42-L aquarium with the test fish, and

vigorously aerated for 30 minutes to facilitate attachment of additional glochidia before a continuous flow of fresh water (18°C) was supplied. Four days after inoculation, the water temperature was raised at a rate of 2°C per day to the final test temperature of 22°C and was maintained at this level for the remainder of the 55-day test. The aquarium was also fitted with a large mesh screen positioned about 35 mm above the entire glass bottom. This screen was used to briefly retain food pellets for the fish while glochidia and/or juveniles that were released from the fish could drop through the mesh screen to the glass bottom and not be ingested. The bottom of the aquarium was siphoned and the fish were offered food pellets at least 3 times per week. Water siphoned from the aquarium first flowed through a 202-µm mesh filter, then through an 80-µm mesh filter. Particles retained by the filters were rinsed with water into petri dishes and thoroughly examined under magnification with dissecting microscopes by two or more individuals to detect sloughed off-glochidia or transformed juveniles. In addition, one or more test fish were anesthetized on days 5, 15, 33, and 51 to examine the gill lamellae and fin tissues for attached glochidia using a dissection microscope. Test fish were euthenized at the conclusion of the test and each gill arch was subsequently excised to thoroughly examine the lamellae for attached glochidia.

No glochidia were observed attached to any part of the fish during the 55-day test. Likewise, no sloughed glochidia or transformed juveniles were observed among the particles recovered from the bottom of the aquarium. However, numerous rotifers were detected in water filtered from the aquarium on day 21 and remained present throughout the remainder of the test. Early life stages of this parasite likely infected the catfish earlier in the year while the fish were being cultured in outdoor ponds and went previously undetected.

Due to the small number of apparently viable glochidia (collected from only one female) that were available to conduct all fish host tests in 2002, and the presence of predacious rotifers in the aquarium during the test, the results of this test should be considered inconclusive. Continued fish host testing is recommended in 2003 under a study design similar to that used in 2002 at the Upper Midwest Environmental Sciences Center, provided that a larger number of viable glochidia are collected from more than one female and additional fish culture practices are taken to limit the possible introduction of unwanted parasites and disease organisms into the test system.