Western Pearlshell Mussel Reproduction in Merrill Creek, Oregon: Timing

2010 Annual Report

Donna Allard, Marci Koski and Timothy A. Whitesel

U.S. Fish and Wildlife Service Columbia River Fisheries Program Office 1211 SE Cardinal Court, Suite 100 Vancouver, WA 98683 360-604-2500



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Introduction

Nearly 300 species of freshwater mussels are native to North America. Nearly three-quarters of these are imperiled as a result of degraded water quality and invasive species (Nedeau et al. 2009, Williams et al. 1993). Within the Pacific Northwest, freshwater mussels were once abundant in the Columbia River Basin but are becoming increasingly scarce (Helmstetler and Cowles 2008). Causes for this decline include dams, water quality impairment, changes in fish populations, introduction of non-native species, and siltation (King County 2005, Nedeau et al. 2009). Freshwater mussels can be an indicator species of ecosystem health because they require clear, clean water in which to live and reproduce (Nedeau et al. 2009). As outlined in the National Strategy for the Conservation of Native Freshwater Mussels (1998), one of the goals is to "increase fundamental knowledge of basic biology and habitat requirements of mussels so that managers can more effectively conserve and manage our mussel fauna." By learning more about the life history of freshwater mussels, conservation and management plans can be developed to assist resource managers in minimizing or eliminating threats and protecting mussel habitat and implementation of these plans will direct the successful conservation of freshwater mussels.

The general reproductive strategy of freshwater mussels has been well documented. During breeding, males release sperm into the water. For fertilization to occur within the marsupium, females must filter sperm from the water. In the marsupium, the embryos develop into larvae called glochidia. Some species such as the western pearlshell (*Margaritifera falcata*) (WPM) release conglutinates, aggregates of glochidia loosely bound by mucus (Barnhart et al. 2008). These conglutinates mimic worms or insect larvae (Figure 1), and rupture if a fish attacks, giving the fish a mouthful of glochidia. Otherwise, the conglutinates disintegrate rapidly, leaving the free glochidia to find a host on their own. The glochidia complete their development as parasites on the gills and/or fins of suitable host fish such as cutthroat trout (Karna and Millemann 1978, Nedeau et al. 2009). In order to survive, glochidia must attach to a suitable host within days to a couple of weeks after being released into the water (Murphy 1942, Jansen et al. 2001). After attached to a host, they form a cyst around themselves and remain attached for several days or months depending upon the species and water temperature (Barnhart et al. 2008). When the glochidia metamorphose into tiny mussels, they release from the fish, burrow into the sediment, and begin their existence as free-living mussels (Hastie et al. 2003).

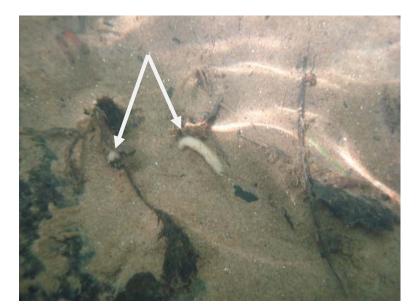


Figure 1. Conglutinates from the Western pearlshell mussel, Margaritifera falcata.

Two critical phases in the life cycle of the freshwater mussel occur: 1) after the glochidia are released and must attach to a suitable host, and 2) when the juvenile mussels are released from the host into the substrate. Mortality of the eastern pearl mussel (*Margaritifera margaritifera*) at these stages was shown to be 99% and 95%, respectively (Young and Williams 1984). In the same study, it was estimated that only one in 1,000,000 glochidia shed from the marsupium survives to become a juvenile mussel. WPM spawning in two western Washington streams appears to be related to water temperature (number of degree-days, Toy 1998). In addition, Eastern pearl mussel spats (the release of glochidia from the marsupium) are likely triggered by an environmental cue such as water temperature or changes in discharge (Hastie and Young 2003, Nedeau et al. 2009). However, in general, information regarding the reproductive timing and triggers of western pearlshell mussel populations is scarce.

The goal of this study was to improve our understanding of WPM reproduction. The specific objectives of this study were to: 1) estimate the WPM population size in the study reach, 2) determine when WPM reproduce in Merrill Creek, Oregon, and 3) determine whether WPM reproduction events can be correlated with an environmental variable (primarily temperature).

Methods

Study Area

Merrill Creek (Oregon, USA) is a 12.7 km long tributary of Tide Creek. It enters Tide Creek approximately 1.8 km upstream from its confluence with South Deer Island Slough (Figure 2). The stream reach we studied is located approximately 3.3 km upstream from the mouth of the creek. The reach is approximately 250 meters in length and on average approximately 5.0 m wide. Mussels are located throughout the reach, both loosely scattered and in patches. Three areas within the study reach with relatively dense populations were identified. The creek in this vicinity is low gradient with a substrate mostly of gravel and sand. A few areas throughout the reach are comprised of cobble.



Figure 2. Study area (indicated by red square in inset) and general location (red dot) of the mussel bed. The Columbia River is northwest of Merrill Creek.

Objective 1: Estimate the population of western pearlshell mussels in the study area

To estimate the number of mussels in the 250 m reach, the entire reach was divided into 250, 1 m wide transects. Mussels were counted in 25 randomly selected transects, representing 10% of the total study area. Every visible mussel was counted within each transect. This represented 10% of the population. To estimate the number of mussels in the reach's population, the average number of mussels in each transect was calculated along with the 95% CI. This transect average was multiplied by a factor of 10.

Objective 2: Determine when western pearlshell mussels are reproducing

Ten permanent transects, each 2 m wide, were established within the reach, and were surveyed throughout the study (i.e., the same transects were surveyed during each field visit) (Figure 3). Three dense areas of mussels were identified in the reach. A dense area of mussels was defined as being approximately 20 m² and having mussels within centimeters of one another throughout the area. Two transects were placed haphazardly in each of the three, densely populated (DP) areas for a total of six transects. One transect was placed above, below, and in between each of the DP areas for a total of four more transects (Figure 3). These transects were added to increase the number of mussels sampled in the population and the likelihood of finding reproductive mussels. At each transect, a total of three mussels were haphazardly chosen and examined for signs of gravidity. Thirty mussels were sampled during each sampling event.

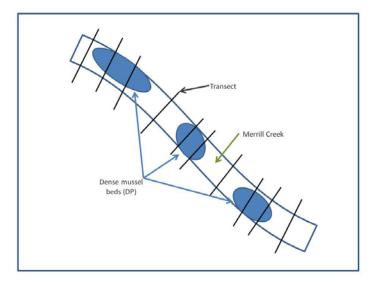


Figure 3. Transect placement throughout the study reach.

Beginning on May 5, 2010, through July 28, 2010, 30 haphazardly-selected mussels were examined once every two weeks. One last sampling date occurred on August 18, 2010. After selection of a mussel, the length of the mussel body that was protruding out of the substrate was measured to the nearest millimeter (Figure 4). Then the mussel was gently removed from the sediment, where a marker was placed at its location in the substrate. The length, height, and width of the mussel were measured with dial calipers to the nearest millimeter (Figure 4). To determine gravidity, the mussel was pried open approximately 2-3 mm using a dull pocket knife and the marsupial gills were examined. A gravid mussel was distinguished by inflated, opaque marsupial gills containing visible patches or striations of eggs or embryos (Spring Rivers 2007, Figure 5). The shell of the mussel was marked with an individually numbered floy tag (Lemarie et al. 2000) and returned to its original location and orientation in the substrate. It was assumed that different mussels would be examined at each survey event; marking the shell allowed us to make secondary observations of survival in the case of resampling a mussel.

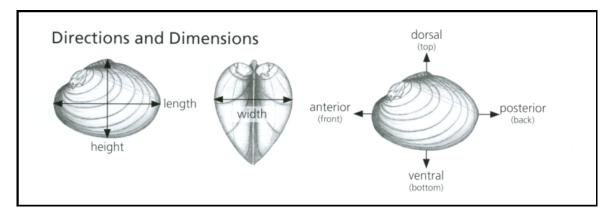


Figure 4. Directions and dimensions of a mussel

During each sampling event the substrate 0.5 m above and below each transect line was examined for the presence of conglutinates using an aquascope. A plankton net (50 μ m mesh) was deployed at the beginning of each sampling event to survey for the presence of glochidia in the creek water. An additional transect that spanned the width of the stream was established at a fixed location 25 m downstream of the study area. Five plankton net samples along the transect were collected during each sampling event, as flow allowed. As velocities along the transect neared 0.01 m/sec the plankton net was not effective, in which case we collected as many samples as flow allowed (Table 2). The plankton net was left in place for ten minutes for each sample. Each sample was preserved in ethanol and taken back to the lab for examination to determine the presence of glochidia.



Figure 5. An example of a gravid mussel's gills (Red arrow points to white opaque gills).

Objective 3: Determine if WPM reproduction events are correlated with an environmental variable (primarily temperature)

On April 8, 2010, two Onset Hobo thermographs were deployed in Merrill Creek. One thermograph was placed in a pool in the lower end of the stream study reach, at approximately 3.5 RKm and the other was placed above the tributary below a fish passage barrier at RKm 8.2 (Figure 2). Previously, a temperature and depth probe was installed at the mouth of Merrill Creek in October of 2009 by the Columbia Soil and Water Conservation District. The probe recorded temperatures once every hour. Several other stream and water measurements were taken just below the stream reach during each sampling event. A discharge measurement was taken using a Marsh McBirney Flo-Mate. Water quality measurements were collected with a YSI 85 meter; these measurements included temperature, dissolved oxygen, and conductivity. A turbidity measurement was taken as well with a Hach 2100p turbidimeter.

Results

Objective 1: Estimate the population of western pearlshell mussels in the study area

To estimate numbers of adult mussels in the reach's population, a total of 239 mussels were counted along 25 transects within the reach, each 1 m wide and on average 5.02 m long. This represented approximately 10% of the 250 m reach. Each transect had an average of 9.6 \pm 4.29 (95% CI) mussels, with a range of 0 to 34 mussels and standard deviation of 10.95. Based on these numbers, we estimated that the reach contained a population of approximately 2400 \pm 1073 (95% CI) mussels.

Objective 2: Determine when western pearlshell mussels are reproducing

The ten transects established to detect mussel reproduction were sampled eight times (Table 1). A total of 208 unique mussels (approximately 8.7% of the estimated population) were examined for gravidity. Of these, 166 were marked with a floy tag (tags were not available until the third sampling event). Eleven (5.3%) mussels were recaptured; one (0.5%) was recaptured twice; and one (0.5%) was recaptured three times. All recaptured mussels appeared to be unharmed from the sampling effort.

Gravidity was not observed in any of the mussels examined during the transect sampling events. However, on May 5, 2010, the first day of sampling, four mussels were observed releasing conglutinates in the upper reach (Figure 5). These mussels were observed within the study reach but not along a transect. Conglutinates were also observed downstream of the mussel spat, on the substrate at this time. These four mussels were not examined (pried open). On May 5, 2010, only 12 mussels were examined. Thirty mussels were examined on all other sample dates. The number of mussels examined is summarized in Table 1.



Figure 5. Four Western pearlshell mussels releasing conglutinates.

The size of the western pearlshell mussels examined in Merrill Creek approximates a normal distribution, with the majority of the mussels falling within the 55 to 75 mm range (Figure 6). Shell length ranged from 32.1 mm to 97.4 mm. Mussels at sizes less than 25 mm were difficult to discern within the substrate and therefore not sampled. The relationship between length and height was linear (Figure 7).

Stream drift samples were collected on six of the eight sampling events. The plankton net was not available until the second sampling event. During the last sample date, August 18, 2010, the discharge of 0.01 CMS was not sufficient to deploy the net. Glochidia were found in the stream drift samples collected during three of the sampling events (Figure 8). The latest observation of glochidia in the samples occurred on June 16, 2010. Table 2 summarizes our plankton net sampling results.

	Number	Number	% Gravid	Conglutinates	
Date	Examined	Gravid	(95% CI)	Present	
5/5/2010	12	0	0 (0-24.3)	Yes	
5/18/2010	30	0	0 (0-11.1)	No	
6/3/2010	30	0	0 (0-11.1)	No	
6/16/2010	30	0	0 (0-11.1)	No	
6/30/2010	30	0	0 (0-11.1)	No	
7/14/2010	30	0	0 (0-11.1)	No	
7/28/2010	30	0	0 (0-11.1)	No	
8/18/2010	30	0	0 (0-11.1)	No	

Table 1. Occurrence of gravid mussels in Merrill Creek, OR

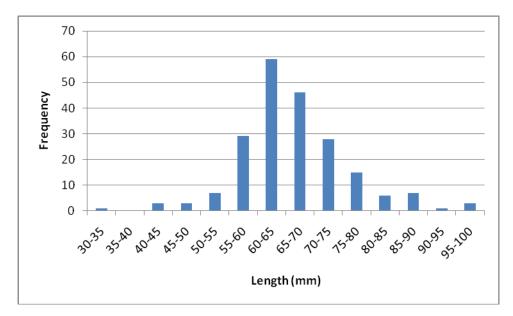


Figure 6. Length frequency of WPM mussels sampled in Merrill Creek.

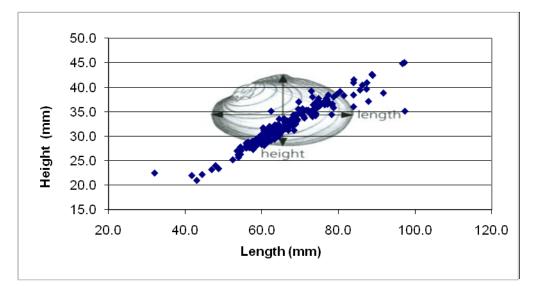


Figure 7. Length vs. height of WPM mussels sampled in Merrill Creek.

Date	Plankton Net Samples	Glochidia Present/#Samples	Discharge (CMS)	
5/5/2010	0		0.20	
5/18/2010	5	Yes/3 out of 5	0.09	
6/3/2010	5	Yes/4 out of 5	0.58	
6/16/2010	5	Yes/1 out of 5	0.23	
6/30/2010	4	No	0.06	
7/14/2010	2	No	0.04	
7/28/2010	1	No	0.02	
8/18/2010	0		0.01	

 Table 2.
 Stream drift sample results.

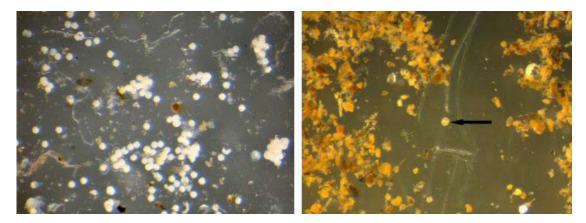


Figure 8. Glochidia collected from spat (left) and collected in stream drift sample (right).

Objective 3: Determine if WPM reproduction events can be correlated with an environmental variable (primarily temperature)

The mean daily temperature in Merrill Creek from April 9, 2010, to September 30, 2010, ranged from a low of 7.5° C on April 9th to a high of 18.4° C on August 17th (Table 3, Figure 9). The mean daily temperature during which glochidia were observed ranged from a low of 8.5° C on May 5th to a high of 13.6° C on May 17th. Glochidia were collected in our stream drift samples until June 16, 2010. The mean daily temperature from May 5th when glochidia were first observed to June 16th was 10.7° C. Other stream and water measurements collected are shown in Table 3 and Figure 10.

Date	Time	Temp (°C)	%DO (mg/l)	DO (mg/l)	Conductivity (µS)	Relative Conductivity (µS)	Turbidity (NTU)	Flow (CMS)
5/5/2010	9:35	7.3	102.7	12.38	36.6	55.3	7.08	0.20
5/18/2010	13:22	13.6	105.5	10.36	48.2	62	8.67	0.09
6/3/2010	10:27	9.7	101.5	11.65	34.3	48.5	9.74	0.58
6/16/2010	8:20	9.8	95.3	10.85	39.4	55.5	6.87	0.23
6/30/2010	9:40	11.5	98.3	10.7	49.2	66.3	4.84	0.06
7/14/2010	10:02	13.1	95.9	10.09	58.9	36.2*	3.85	0.04
7/28/2010	9:43	14.9	89.4	8.9	67.8	83.9	4.84	0.02
8/18/2010	9:10	16.4	78.2	7.64	80	95.8	3.44	0.01

Table 3. Water quality and stream discharge measurements.

* Data Point probably recorded incorrectly.

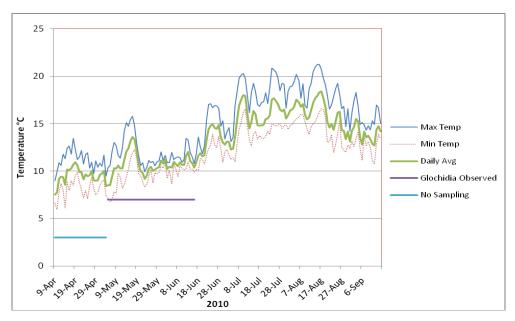


Figure 9. Maximum, minimum, and mean daily water temperatures in Merrill Creek from April 9th to Sept. 30th, 2010.

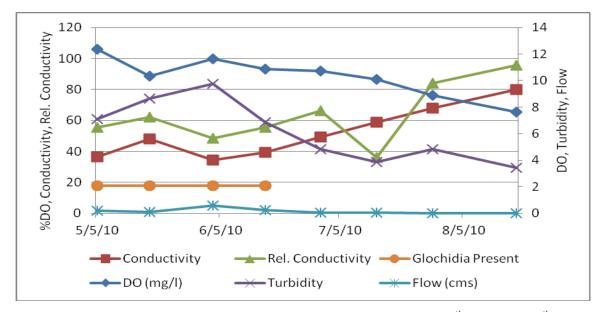


Figure 10. Stream and Water Measurements in Merrill Creek from May 5th to August 16th, 2010.

Findings

- 1. The estimated population size (> 30 mm) studied was approximately 2,390 WPM. We examined a total of 208 mussels. Given that freshwater pearl mussels generally do not reach sexual maturity until an age of approximately 15 years (Bauer 1987, Skinner et al. 2003) at which time they generally reach a shell length of approximately 50 mm (Toy 1998), we estimate that 90% of the mussels in this reach (2,151) could have been reproductive (187 of those sampled). If we assume a 1:1 sex ratio (Young and Williams 1984, Toy 1998, Spring Rivers 2007, King County 2005), and that we were equally likely to sample males and females, we would have sampled approximately 94 potentially reproductive females, or approximately 8.7% of the potentially reproductive female population (1,078). If we assume that as many as 35% of the potentially reproductive females are not reproducing in a given year (Bauer 1987, Spring Rivers 2007), we would estimate that a maximum of 701 of the mussels in the reach and 61 of the mussels we sampled may have been reproductive this year. Thus, although the sample size of reproductive females may have been as low as 2.6% of the population, our sample design was reasonable to detect gravidity. However, no WPM examined (pried open) were suspected of being gravid.
- Evidence of WPM reproduction was found from May 5, 2010 (1st sample date) through June 16, 2010 by either observing conglutinates or finding glochidia in drift samples.

- Reproduction was coincidentally detected in four mussels on May 5th, 2010. These animals were not part of our study group and were not examined. The mean daily temperature on that day was 8.5°C (range 7.0-10.0°C).
- When evidence of WPM reproduction occurred, the average daily temperature was 10.7° C, and the maximum daily temperature exceeded 10°C.
- When no further evidence of WPM reproduction was found, the minimum daily temperature exceeded 10°C.
- 6. Signs of reproduction were not detected in any of the 208 selected animals.

Conclusions

Other studies on the pearlshell mussel have suggested varying times of reproduction and glochidial release, between and within rivers, that may be dependent on water temperature. Studies have also indicated glochidial releases may coincide with flood events, possibly resulting in sedimentation and interfering with mussel respiration (Hastie and Young 2003). With only one season of field study, it is difficult to determine any correlation with environmental variables such as temperature or discharge.

This report details the results of a first year pilot study. It would be beneficial to continue this study in future years to gain a better understanding of the role certain environmental variables, especially temperature and discharge, may play in the timing of WPM reproduction. Since the observation of WPM spats were observed on the first day of field sampling, work should be scheduled to begin no later than April. If a similar small stream with WPM is identified in the area, a comparison of mussel reproductive timing and temperatures would also be useful.

This study is a small part of a bigger effort of research taking place on Deer Island. The information gathered will be integrated with information about other species and habitat use on Deer Island that we are collecting. Using the Strategic Habitat Conservation approach, this combined information may help determine what restoration activities, if any, will benefit the watershed in the future.

Acknowledgements

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