PARTNERSHIP FOR THE DELAWARE ESTUARY

Re-Introduction of Freshwater Mussels into Red Clay and White Clay Creeks, DE

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Interim Report for Delaware Clean Water
Advisory Council

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Established in 1996, the Partnership for the Delaware Estuary is a non-profit organization based in Wilmington, Delaware. The Partnership manages the Delaware Estuary Program, one of 28 estuaries recognized by the U.S. Congress for its national significance under the Clean Water Act. PDE is the only tri-state, multi-agency National Estuary Program in the country. In collaboration with a broad spectrum of governmental agencies, non-profit corporations, businesses, and citizens, the Partnership works to implement the Delaware Estuary's

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Introduction

Freshwater mussels are the most imperiled of all the flora and fauna across North America, as well as in the State of Delaware. In the Delaware River Basin, and specifically northern Delaware, approximately 12 species of freshwater mussels are considered native. However, only one of these species has been found in the last ten years in any numbers above the head of tide in the Christina Basin, which extends from Wilmington to the west and north into Pennsylvania and includes the Brandywine, Christina, White Clay and Red Clay tributaries.

The decline of the freshwater mussel assemblage includes reduced biodiversity, population sizes, and extent. Many streams no longer have any mussels of any species, and when mussels are found they have been of only the two most common species (*Elliptio complanata* and *Pyganodon cataracta*). Furthermore, it is unclear if the remaining vestigial mussel populations are successfully reproducing since there have not been any documented sightings of juvenile mussels anywhere in the state in recent decades, based on data available to the Partnership for the Delaware Estuary (PDE).

Historical data are limited, but there is ample evidence to suggest that major streams in northern Delaware once harbored diverse and robust mussel assemblages. In his surveys of Pennsylvania streams that drain to the Christina, Ortman (1919) recorded up to seven species of mussels in the upper White Clay, Red Clay and Brandywine Creeks, and typically mussel densities increase in lower portions of rivers. Based on studies of reference streams elsewhere, the natural mussel groupings would have consisted of aggregated populations of several species, occupying different niches (benthic habitats) within the stream, and collectively filtering a tremendous amount of water.

Reasons for the Decline. Mussels likely become extirpated from streams because of either: 1) general impaired water or habitat quality, 2) specific incidents (i.e. spills) that cause acute mortality in a single event, 3) overharvesting/predation, and/or 4) loss of fish host species to support larval mussel growth and distribution. Once extirpated from a stream or reach, mussels are not able to recolonize easily because either there is no longer broodstock nearby, or dams and other impediments to fish passage may block dispersal of juveniles (via fish hosts) back into the stream. Mussels have a long lifespan (30-75 years or more) and do not reproduce until at least 8 years old. Therefore, even if water quality is improved and conditions permit redistribution via fish hosts, it can take decades for natural recolonization and recovery. Over time, the basin's metapopulation has become increasingly fragmented and less resilient. Over the past 10 years, scientists from PDE and partners have surveyed more than 70 streams in the area and mussels were found in only 4 streams.

The life cycle of freshwater mussels is very different from marine bivalves because their reproduction is dependent on migratory fish hosts whose journey upstream is often impeded by dams. Dams also harm mussels by altering habitat and flow conditions. Our discoveries of extant beds of several rare species in 2009 and 2010 occurred in the undammed main stem Delaware River. Although efforts to remove dams on the tributary streams have been accelerating, it will be some time (maybe decades) until fish hosts for freshwater mussels will range freely and help to re-disperse mussels back into the streams from the remaining beds on the tidal Delaware River. Furthermore, even if dams are removed and mussel juveniles recruit into those streams, it will take decades until the mussels grow large enough to begin to propagate on their own. In the meantime, the diverse remnant beds in the tidal Delaware River will be vulnerable to spills or other impacts associated with life in an urban corridor.

The Freshwater Mussel Recovery Program. Rather than wait for natural populations to expand, which may or may not occur, we aim to assist in dispersing mussels throughout suitable streams in their historic range to promote resilience of the remaining, vulnerable metapopulation. Ultimately, we would like to strengthen and hasten mussel restoration efforts by propagating seed juveniles in a hatchery for use in restocking. PDE has led efforts in the Delaware River Basin to develop hatchery propagation methods and facilities, but due to limited funding and capacity those efforts might take some time to reach fruition. In the meantime, PDE is leading efforts to assess which streams should be prioritized for mussel restoration, beginning with incremental restocking by relocating reproductive adults from healthy populations. We have also been actively working to raise awareness about the importance of mussels and their conservation and restoration via outreach to the public and elected officials. The Recovery Program consists of various activities described on the PDE website: http://www.delawareestuary.org/science_projects_mussel_restoration.asp.

Benefits of Mussels. Diverse and robust mussel beds help to sustain healthy streams and rivers by improving water quality, enriching the ecology, and providing one of the best possible bioindicators of stream health for environmental managers. Our research has shown, for example, that the relic population of mussels in one 6-mile stretch of the Brandywine River filters more than 25 tons of suspended, polluting particles per year. Each adult mussel can filter up to 15 gallons of water per day, thereby helping to remove algal pollution fueled by excess nutrients, resulting in improved light conditions for healthy bottom plants. Increasingly across the United States and world, bivalve shellfish restoration is being used to help remediate degraded water quality.

Therefore, while we are interested in promoting native biodiversity the principal focus of our mussel recovery efforts is to restore a natural mixed-species assemblage of freshwater mussels for water quality improvement via NPS pollution reduction. Our efforts to date have focused on refining propagation techniques for mussels, selecting streams for mussel restoration, reintroducing mussels to targeted streams via relocation methods, and building awareness through outreach programs. Where feasible, we have also developed pre-restoration testing methods to identify which streams are capable of sustaining mussels so that these various recovery efforts target streams where success is more likely. We have mainly been implementing the program with two common mussel species to first stabilize and expand their ranges, but in the future we aim to expand the effort to include other native mussel species so that restored mussel assemblages are more resilient and functional than just a single species.

Project Context with the FMRP and Goals. Initial efforts to develop and launch the Freshwater Mussel Recovery Program (FMRP) have been undertaken within the Commonwealth of Pennsylvania mainly because of PA sources of funding, more available survey data, and high interest from state agency biologists. Within Pennsylvania, we have had success with each of the five FMRP activities (surveys, stream suitability tests, reintroduction trials, hatchery propagation and outreach). ***The goal of this study was to launch pilot FMRP activities within the State of Delaware by performing surveys and a reintroduction trial within the Christina Basin***. Our specific objectives will be to:

- Confirm the viability of mussel sources on the lower Brandywine River.
- Perform historical data review and new mussel surveys to confirm the restoration need within the lower White Clay Creek (expanded to include Red Clay Creek with a Clean Water grant)
- Perform a pilot reintroduction trial whereby up to 200 adult mussels are relocated from the lower Brandywine River to the lower White Clay and Red Clay Creeks, and thereafter monitor them for 1 year using electronic tracking.

- Expand the Freshwater Mussel Volunteer program into the state of Delaware
- Estimate the potential water quality benefits of expanded mussel restoration in White Clay and Red Clay Creeks.

If successful and contingent on funding from other sources, PDE and partners would aim to expand the FMRP within the State of Delaware in various ways (more streams, more species, more animals, more quantitative water quality goals). Additional information about the FMRP is available upon request.

Study Methodology

Study Creek Description

The Red Clay and White Clay Creeks originate in southeastern Pennsylvania and flow through northern Delaware after which they meet at their confluence and flow into the Christina River. The Red Clay Creek, which is considered a tributary of the White Clay, has a total watershed area of 17,212 acres which is substantially smaller than the White Clay Creek watershed (31,647 acres). Red Clay creek watershed encompasses the towns of Kennett Square PA, known for its mushroom farms, and Stanton, DE. The White Clay watershed encompasses a large area in rural southeast PA, including the boroughs of West Grove and Avondale and the suburbs of New Castle County, DE. Both creeks are characterized with a sandy bottom with riffle areas consisting of bedrock and other hard substrate. Both streams are considered impaired by stormwater runoff.

Qualitative Stream Surveys

During the spring of 2013, PDE field crews surveyed both Red Clay and White Clay Creeks to determine presence of freshwater mussels in each creek.

A qualitative time-search approach was adopted to enable large areas to be searched using best available methods, generally wading with viewing buckets and polarized glasses. Surveys were characterized by searching for any evidence of mussels (shell valves, live mussels, etc.). Surveyors spread across the creeks and searched on both banks as well as near riffles, in pools and along areas of good mussel refugia.

Figure 1. Jessie Bucker, Angela Padeletti and Kurt Cheng (left – right) surveying the White Clay Creek.

All mussel surveyors were trained and experienced, and for comparative purposes their search results can be standardized as “catch-per-unit-effort” whereby effort is measured in “person-hours” and reach length traversed. Qualitative surveys were performed by 2-3 field personnel at any one time (Fig. 1) and ranged from the Delaware state line to the confluence of the creeks as seen in Figure 2.

Surveys were designed to be representative of the respective creek as it was not possible to survey the entire creek length within the scope of the project. Mussel shells typically wash downstream once a mussel dies and are easily found in shallow areas and on exposed bars. If mussel s existed between survey reaches, signs of their existence would likely have been detected within the search areas. Therefore, we consider this approach and coverage representative of prevailing conditions.

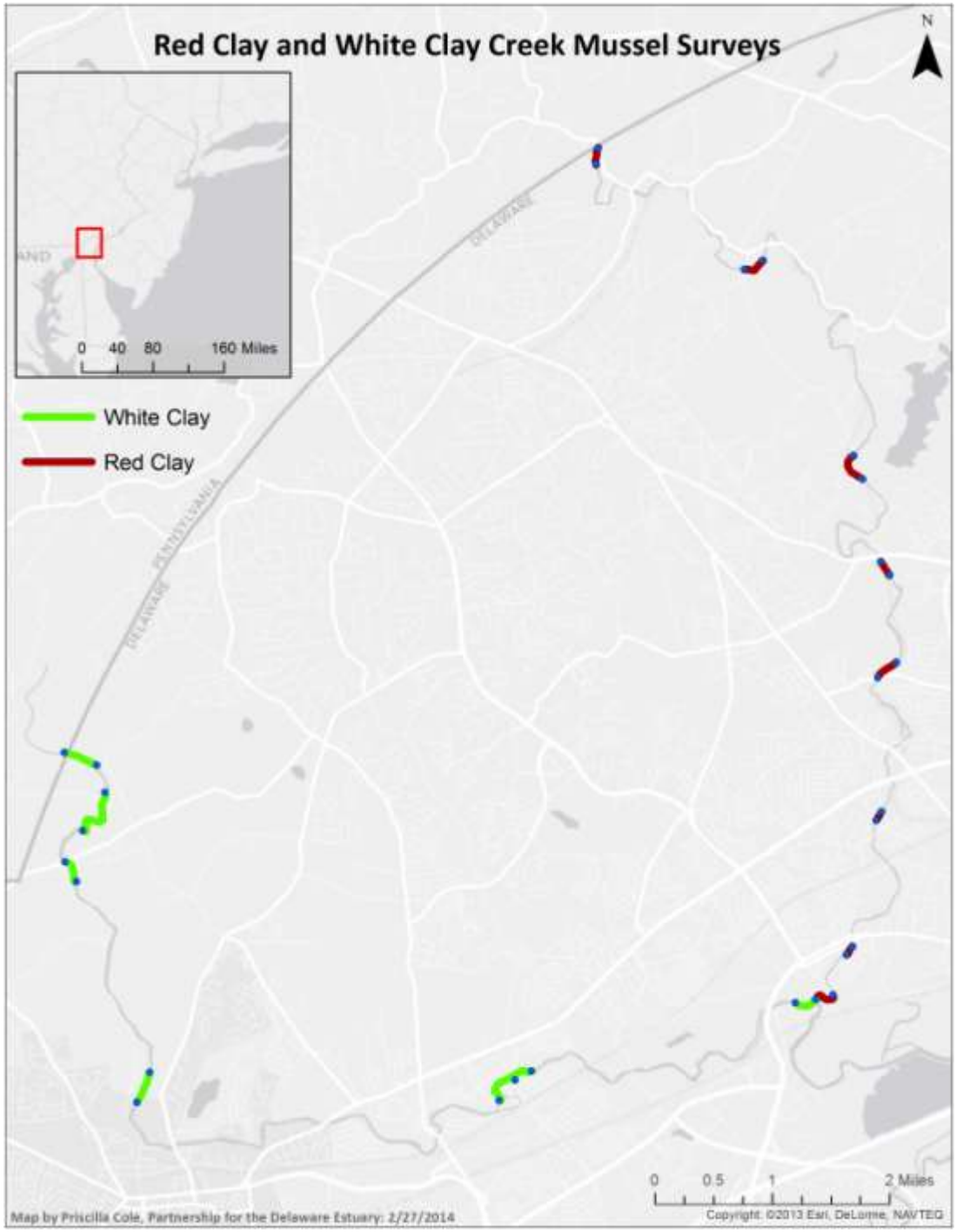

Figure 2. Locations of qualitative stream surveys performed by PDE in the Red Clay and White Clay creeks. Surveys went from the Delaware State line to the confluence of the two creeks.

Quantitative Survey of Extant Population

PDE performed a quantitative survey at one example location of the lower Brandywine River within Delaware on November 21, 2012, to assess whether high enough numbers of live *Elliptio complanata* still exist (e.g., >10,000 adults) to sustain mussel restoration efforts. Kreeger had earlier performed an intensive mussel survey of the lower Brandywine River in Pennsylvania in 2000, and so our 2012 goal was simply to add an additional sampling station in Delaware to contrast with the extensive earlier dataset. PDE also performed qualitative mussel surveys (timed searches) in random stretches of White Clay Creek.

Thompson's Bridge was chosen as the area to survey in the Brandywine because large numbers of mussels had been observed in this area by PDE science and outreach staff. Two areas were surveyed, one above the bridge (4,686 m^2; Fig. 3), and a smaller area below the bridge (798 m^2). Both areas were along the left descending bank in water less than 2 feet deep.

Figure 3. Approximate mussel survey area upstream of Thompson's Bridge, DE.

The Brandywine quantitative survey consisted of 12 transects, each extending perpendicular to the river flow. Up to three quadrats (each 1 m^2; aluminum frame) were randomly placed along each transect, extending from near bank to 21 m from the left descending bank. Water depths were up to 2 feet deep. Quadrats were searched visually using polarized glasses and a viewing bucket. Live mussels were removed with tongs and placed into collecting bags. Following a visual search, a scoop sieve (1 cm mesh) was used to search the sediments up to 10 cm deep within the entire quadrat. Sieve searching typically revealed additional animals that were apparent visually, especially since the colder late fall waters can lead to mussel burial. Sieve searching is also the only effective method for locating juvenile mussels. Collected live mussels were taken to shore for species identification and recording of shell heights with digital calipers (±0.01 mm), and then were replaced in the stream bottom. GPS readings were taken within each quadrat for mapping of the mussel bed and delineation of the survey area. Surveyors were Danielle Kreeger, Joshua Moody, and Doug Janiec.

Mussel Relocation

For this project, mussels were first collected from the Thompson's Bridge site on the Brandywine River and they were promptly dual-tagged. Following tagging, similar size ranges were apportioned to replicate groups that were transported to various locations in White Clay and Red Clay Creeks. Subsequently, mussels were surveyed using an electronic tag reader (explained below) to determine how well mussels were retained at the release locations in each stream. The dates for these milestone activities are summarized in Table 1.

Date (2013)	Activity
August 15th	Native Mussel Collection
August 16th	Mussel Deployment
November 16th	Reintroduction Survey

Table 1. Summary of re-introduction steps

Collection. On August 15th, a field crew of four mussel collectors waded the Brandywine River at the Thompson's Bridge mussel bed (see above) to collect a total of 254 *E. complanata* specimens using collecting bags. Mussels were then transported to shore where they were cleaned using soft brushes and river water.

Tagging. Once shell surfaces had dried by air exposure on tarps, mussels were dual-tagged with a unique numbered plastic tag as well as a unique numbered electronic passive integrated transponder (PIT) tag. Plastic tags were affixed to the shells of mussels via super glue. PIT tags were encased in marine epoxy. The plastic tag served to easily identify mussels visually. The PIT tag allows for mussels to eventually be detected in streams without the need to pick them up and disturb them. Each PIT tag number is unique and was recorded by an electronic tag reader.

Once tags were properly applied and dried onto the mussels to be relocated, tag numbers were recorded and each mussel's shell height was measured with digital calipers (±0.01mm). Recording shell heights allows researchers to assess growth rates of relocated mussels (later). This provides an understanding of the habitat quality and food conditions within the relocation streams.

After shell heights and tag numbers were recorded, mussels were then divided into seven groups of 30 individuals. A group of 44 additional mussels were separated to be placed into the Newark Reservoir as an additional relocation effort (not connected to any grant, but reported here). Each group had a similar array of mussel sizes from small to large. Each group of mussels was then placed into a collecting bag, and all bags of mussels were then held in the Brandywine River overnight prior to deployment on the next day into Red Clay and White Clay Creeks (as well as the Newark reservoir). These tasks are depicted in Figure 4.

Deployment. On the 16th of August, PDE personnel deployed the various groups of mussels into Red Clay and White Clay Creeks and the Newark reservoir. The locations of the deployment sites are shown in Table 2. Locations were chosen based on observations during earlier mussel surveys where we identified places that have suitable habitat for mussels based on prior experience. Suitable

Figure 4. (Top-Bottom) Plastic tag being applied to a mussel with super glue. PIT tag being applied to a mussel with marine epoxy. Gus Wolfe using a PIT-tag reader to record the PIT tag number of a mussel. Grouping of mussel to ensure similar size ranges per bag by Dee Ross (left) and Kurt Cheng (right).

habitats generally are along stream banks in moderate depths where the substrate appears stable and less prone to stormwater driven bed transport during flooding events. Mussels prefer course substrate that they can bury into, but which does not move during flooding. This approach has proven successful in earlier relocation studies in PA, with mussels surviving flooding during Hurricanes Irene and Sandy, in previous relocation tests. Mussels were hand placed with anterior ends pointed down into suitable sediments within each relocation area (Fig. 5), to create small mussel beds of 10-15 mussels per site, and a few sites per location. Deployment teams recorded coordinates of deployed mussel beds for future reference and to aid future PIT tag retention surveys.

Location	Latitude	Longitude	Deployed
Red Clay 1	39.73005	-75.63648	30
Red Clay 2	39.74777	-75.63631	30
Red Clay 3	39.80888	-75.68077	30
White Clay 1	39.69105	-75.75406	30
White Clay 2	39.70516	-75.75668	30
White Clay 3	39.73321	-75.75995	30
White Clay 4	39.73658	-75.76133	30
Newark	39.69344	-75.74435	44

Table 2. Locations where tagged *Elliptio complanata* were deployed.

Figure 5. Angela Padeletti deploying tagged mussels in Red Clay Creek (left). A deployed mussel in creek bed with PIT tag showing (right).

Retention Survey

On the 16th of November 2013, PDE field personnel surveyed the seven deployment locations in Red and White Clay Creeks, as well as the Newark reservoir, to determine whether or not mussels were still present in the immediate areas where they had been released. This was the first of what we expect to be numerous return visits at the locations. During the 3-month period between release and monitoring, major flooding had occurred in these streams.

Due to budget constraints, surveying with a PIT tag reader was only performed within the immediate reach where mussels had been released. If a mussel had become dislodged and was washed downstream, we would therefore not have detected it at the immediate release location. Since we therefore do not know the fate of any mussels that disappeared (e.g. mortality versus simply washing

into a new place where they survive), we refer to mussel repeat finds as "retention" within the immediate release location. Typically, survey areas are about 40-50 meters long and 15-30 meters wide. Our first "retention survey" was therefore performed exactly 3 months after the deployment. Surveying was performed by one person who carefully waded through the release location using a PIT-tag reader with an extended antenna, as shown in Figure 6. The antenna must pass within 18 inches of a tag to register a hit. Therefore, it is very important that the surveyor use slow sweeping motions with the antenna in a rectangular, overlapping sweep motion to avoid missing animals. The reader was also held just above the creek bottom in order to scan for any deployed mussels.

Figure 6. Danielle Kreeger surveying a deployed mussel bed using an FS-2001 PIT-tag reader.

When the PIT-tag reader scans a PIT tag, the reader emits an audible beep and the unique tag number is recorded and displayed on the reader's screen. A second person wading beside the main surveyor assisted with note taking and monitoring safety. In cases where a longer time has passed since deployment, the second person would also pick up animals and measure their shell heights and then redeploy them, but the 3-month period in this case was too short to detect significant new shell growth.

Survey efforts were distributed equally among the various locations. For the purposes of this first monitoring visit, we did not spend as much time at each site as we would have liked, and so we likely missed some animals, but since all sites were surveyed similarly, relative differences seen in the retention data should be reflective of differences in site performance. Typically, we will survey each deployment location in depth once per year, where we spend longer time and we size every animal we encounter.

Volunteer Mussel Survey

Fifteen mussel trainees attended the Delaware Freshwater Mussel Volunteer Survey (FMVS) workshop on May 18, 2013 at the Brandywine Creek State Park. After a 90-minute classroom introduction to freshwater mussels, survey training program and a safety overview, the group headed to the Brandywine River to get a hands-on lesson in searching for freshwater mussels. Using bathiscopes, clear-bottom buckets, and clear plastic cake pan lids, all survey trainees were able to find mussels in the stream, and by using the Freshwater Mussels of the Delaware Estuary Guidebook, identified them as Elliptio complanada - the most commonly found mussel in the Delaware Estuary. All workshop participants were able to take home handout materials and Guidebooks, and prepared to conduct mussel surveys on their own in other Delaware Estuary streams. Those who were able uploaded their findings (or lack thereof) to the data portal on www.DelawareEstuary.org created for this purpose. The Delaware Nature Society utilized this training to lead a volunteer survey of over 2 miles in the Red Clay Creek watershed. No mussels were located, but stream conditions were recorded in the data portal.

On September 6, 2013, PDE staff (Dee Ross and Cheryl Jackson) presented the FMVS program to the Christina Basin Task Force stormwater bus tour. 40 people met streamside for a first-hand look at the mussels of the Brandywine River, and learned how mussels help clean our waterways and how they

Figure 7. A child shows off the mussel he found while exploring the Brandywine River (left). Dee Ross of PDE, speaks to people from the Christina Basin Task Force about the Freshwater Mussel Volunteer Program at the Brandywine River.

could become volunteer survey leaders. Among the attendees were several organization representatives who have become partners in PDE's freshwater mussel work.

Project Results

Qualitative Survey Results

Red Clay Creek was surveyed a total of 3 times including May 15th, July 16th and July 18th. Combining the three survey days, 1.2 representative stream miles were surveyed in Red Clay Creek. White Clay Creek surveys were completed over 2 field days including the 15th and 21st of May. Nearly 2 representative stream miles were surveyed during those two days. Survey data are summarized in Table 3. **The overall stream miles surveyed for both Creeks amounted to 3.2 miles.**

Survey	Date	Stream Miles	Total Miles for Creek
Red Clay Creek Survey 1	May 15th	0.209	
Red Clay Creek Survey 2	July 16th	0.773	
Red Clay Creek Survey 3	July 18th	0.386	1.158
White Clay Creek Survey 1	May 15th	0.962	
White Clay Creek Survey 2	May 21st	1.027	1.989

Table 3. Total stream miles surveyed on each survey day in Red Clay and White Clay Creeks

Surveyors did not find any evidence of mussel presence (e.g. live mussels, dead shells or valves) in either creek. Despite the lack of freshwater mussels, surveyors observed good mussel habitat and healthy fauna nearby, suggesting that mussel recovery efforts might be successful and that the creeks appeared capable of supporting mussels based on qualitative observations (Fig. 8).

Figure 8. Stream reach of Red Clay Creek (left). Snapping turtle in Red Clay Creek (right)

Quantitative Survey Results

Mussel abundances in the Brandywine per quadrat are shown in Table 4. Quadrat depths ranged between 6-23 inches (0.15-0.58 m). Maximum mussel density per square meter was 33 and the minimum was 0. The average mussel density above Thompson's Bridge was 7.4 mussels m^{-2} and the average below the bridge was 11.5 mussels m^{-2}.

Area	Transect	Quad	Waypoint	Depth (inches)	Depth (m)	Mussels
1	1	1	9	14.5	0.3683	28
1	1	2	10	17.5	0.4445	9
1	1	3	11	7	0.1778	3
1	2	1	15	7	0.1778	28
1	2	2	17	6.5	0.1651	0
1	2	3	18	13.5	0.3429	1
1	3	1	20	8	0.2032	28
1	3	2	22	11.5	0.2921	0
1	3	3	23	6	0.1524	0
1	4	1	26	17	0.4318	1
1	4	2	27	14	0.3556	6
1	4	3	28	18.5	0.4699	1
1	5	1	31	18.5	0.4699	2
1	5	2	32	17.5	0.4445	4
1	5	3	33	18	0.4572	0
2	7	1	35	23	0.5842	10
2	8	1	38	20	0.508	9
2	9	1	40	20	0.508	8
2	10	1	42	18	0.4572	33
2	11	1	44	21	0.5334	1
2	12	1	47	22.5	0.5715	8

Table 4. Mussel abundance per sampling quadrat and quadrat depth along transects within the mussel bed above (area 1) and below (area 2) at Thompson's Bridge, Delaware.

Brandywine survey area 1 above the bridge started at the uppermost end of an island (39.81948797°, -75.57148199°) and proceeded downstream along the left descending bank, between the island and shore, and continued downstream toward Thompson's Bridge following the same bank. The width of this survey belt ranged between 8-21 m, and the length was 309.5 m, corresponding to a total area of 4,686 m^2. Survey area 2 was below Thompson's Bridge along Brandywine State Park, extending 266 m long and up to 3 m wide, for a total survey area of 798 m^2 (ending lowest at 39.81495998° -75.56864597°). Therefore, the total area of the two polygons searched was 5,484 m^2.

A total of 180 mussels were sampled in the Brandywine and their shell height was recorded. One hundred and eleven mussels were found in Survey area 1, while sixty-nine mussels were found in Survey area 2. The shell height ranged from 45.98mm to 90.56. Shell heights did not differ significantly between Survey area 1 (Transect 1-6) and Survey area 2 (Transects 7-12), Figure 8. Figure 9 shows the shell height distribution of all animals. Of note is the distribution of both young and old animals.
To estimate the total number of mussels living within the surveyed area above Thompson's bridge, the mean density (7.4 m^{-2}) was multiplied by the surveyed area (4,686 m^2), yielding a total of 34,675 mussels. The number of mussels living within the surveyed area below the bridge (density of 11.5 m^{-2} x surveyed area of 798 m^2) was estimated to be 9,177 mussels. The total area surveyed (5,484 m^2) is therefore estimated to contain 43,852 mussels.

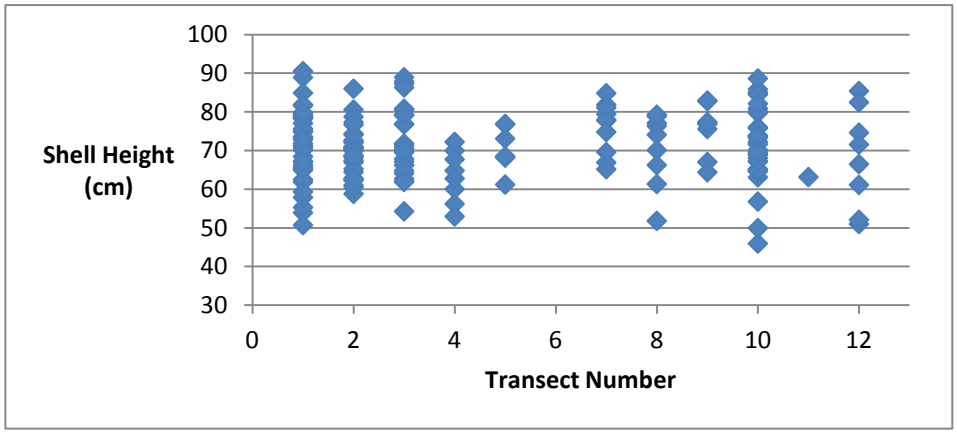

Figure 9. Shell height distributions per transect. Transects 1-6 were found in Survey area 1, and transects 7-12 were in Survey area 2.

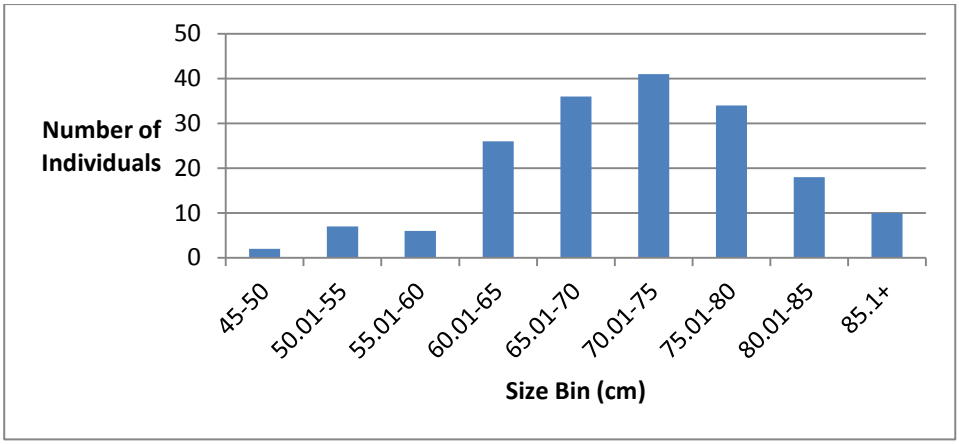

Figure 10. Shell height distribution found in all Survey areas.

Retention Survey Results

Retention data at the various sites in Red and White Clay Creeks are summarized in Tables 5 and 6. Bed retention for deployed mussels differed by location within each of the creeks. Red Clay 1 had less than half of the bed retention than both of the other Red Clay locations. Similarly, bed retention varied at all locations in White Clay Creek.

Location	Species	Number Deployed	Number Recovered	Bed Retention
Red Clay 1	*Elliptio complanata*	30	13	43%
Red Clay 2	*Elliptio complanata*	30	27	90%
Red Clay 3	*Elliptio complanata*	30	25	83%
White Clay 1	*Elliptio complanata*	30	29	97%
White Clay 2	*Elliptio complanata*	30	21	70%
White Clay 3	*Elliptio complanata*	30	17	57%
White Clay 4	*Elliptio complanata*	30	14	47%
Newark Reservoir	*Elliptio complanata*	44	21	48%

Table 5. Retention of freshwater mussels at each deployment location.

Location	Species	Number Deployed	Number Recovered	Bed Retention
Red Clay Creek	*Elliptio complanata*	89	65	73%
White Clay Creek	*Elliptio complanata*	119	81	68%
Newark Reservoir	*Elliptio complanata*	44	21	48%

Table 6. Summary of freshwater mussel bed retention for each deployment creek.

Discussion

Qualitative Survey

Our qualitative survey results indicated no presence of freshwater mussels in the stretches surveyed for either Red Clay or White Clay Creek. While there is a possibility that freshwater mussels do exist within both of these creeks, we surveyed what appeared to be the most habitable areas based off of aerial photography and personal knowledge of the area. Surveys indicated that there was suitable habitat in both creeks for freshwater mussels. Surveys also concluded that both creeks have potential habitat complications with high storm water runoff causing the incising of streams and lack of riparian buffer in certain areas. These observations served as a baseline for freshwater mussels in these creeks and justified our relocation project.

Quantitative Survey

These results confirm that mussels are abundant within at least some areas of the lower Brandywine River within the State of Delaware. More than 40,000 mussels were estimated to reside within an approximately 0.5 km long reach that we refer to here as the Thompson Bridge Mussel Bed. Furthermore, our survey only covered shallow areas along the left descending bank. Mussels are likely to also be present in the middle (deeper areas) and right descending bank of this section of the river, a much great area since the river is approximately 60 m wide in this stretch. Mussels were evident above and below the surveyed reach too, and the team did not attempt to find the upper and lower extent of the Thompson Bridge Bed. It is reasonable to extrapolate that >100,000 mussels exist within the 1-mile reach above/below the bridge. This finding is consistent with the earlier survey upstream in Pennsylvania (Kreeger, summer 2000) where >500,000 *E. complanata* were estimated to reside within the lower 6-mile reach of the Brandywine River in Pennsylvania. Indeed, more mussels may reside in Delaware because the mean density at Thompson's Bridge was more than twice (8.57 mussels per square meter) the maximum density recorded for any intensively surveyed bed in Pennsylvania (3.67 mussels per square meter).Freshwater mussels grow slowly and live to be quite old (up to 80-100 years), and they do not become reproductive until 8-10 years old. Nevertheless, the removal of mussels for the purposes of scientific research or restoration is likely to be insignificant to the viability of the population so long as the population is at least somewhat active reproductively and the removal represents <1% of the population size per year. Mussel fecundity is generally high if suitable fish hosts are present. Although not abundant, numerous large juvenile mussels were found in the current survey at Thompson's Bridge, and a healthy size range was evident.

Based on these results, we concluded that that up to 400 *E. complanata* could be safely removed from the Thompson's Bridge mussel bed per year for scientific and restoration purposes without significantly impacting the viability of the mussel population. Since this is one of the last places to find live mussels in northern Delaware, the very existence of the taxa is at risk by having all animals in only a few places. By helping to disperse a few of these animals as seed populations in other streams where they once lived, it should help strengthen the resilience of the overall metapopulation by broadening their range,

helping to ensure that some survivors would remain If a spill or other local disturbance event depleted the main beds.

Our recommended relocation numbers are conservative because the actual mussel abundance at Thompson's Bridge is certain to exceed the 43,852 animals estimated within the immediate survey polygon. Since freshwater mussel populations are in decline in many areas for reasons that are not fully understood, we further recommend that the Thompson's Bridge bed be periodically monitored to confirm that it remains healthy (e.g. resurveyed every few years).

Retention Survey

Interestingly, the average overall bed retention for Red Clay and White Clay Creeks was similar, about 70%. This was considered very successful and reassuring because of major flooding that occurred during the 28th of August, 22nd of September and 11th of October where stream levels were over 5 times their average flow rates, not long after deployment. As noted above, our November survey was rapidly performed (due to weather constraints at the time). In previous relocation studies, we have had occasions where we subsequently see increases in bed retention following surveys where we don't have enough time to double sweep the release locations to ensure we detect all animals present.

Taken together, these early results indicate that freshwater mussels released into White Clay and Red Clay Creeks are surviving despite some stormwater disturbance events. Longer term monitoring is needed to see whether this success continues, which would ultimately be confirmed if we find positive shell growth after 1-2 years. The long-term desired outcome would be to find juvenile progeny in the area.

Future Research

Further funding is currently being sought to sustain the long-term monitoring of the relocated populations and to expand the survey area for extant populations to include some potential refugia within these drainages that might have been invisible to our stream surveyors (e.g. farm ponds connected to small tributaries). Future PIT tag surveys would enable data to be collected on shell growth (a sublethal condition indicator) as well as bed retention (acute indicator). Since mussels grow slowly and sublethal effects take a longer time to become apparent, subtle differences in streams and reaches can be discerned best by tracking such sublethal measures, thereby guiding where to strategically invest in any expanded restoration efforts for freshwater mussels.