



# Illinois Department of Natural Resources

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www.dnr.illinois.gov

Bruce Rauner, Governor  
Wayne A. Rosenthal, Director

## AMENDMENT TO INCIDENTAL TAKE AUTHORIZATION ISSUED TO THE ILLINOIS DEPARTMENT OF TRANSPORTATION AND THE IOWA DEPARTMENT OF TRANSPORTATION ON JULY 15, 2016 (IDNR ITA FILE #56)

### **Background:**

The Illinois Department of Transportation and the Iowa Department of Transportation (hereinafter referred to collectively as DOT) with Stantec Consulting Services prepared a conservation plan as application to the Illinois Department of Natural Resources (hereinafter referred to as Department) for the incidental take of Higgins eye (*Lampsilis higginsii*), spectaclecase (*Cumberlandia monodonta*), sheepnose (*Plethobasus cyphus*), butterfly (*Ellipsaria lineolata*), ebonyshell (*Fusconaia ebena*), and black sandshell (*Ligumia recta*) mussels during activities associated with the Interstate 74 (I-74) Bridge replacement across the Mississippi River between Moline, Illinois, in Rock Island County, and Bettendorf, Iowa, in Scott County. The conservation plan was received by the Department on May 5, 2016. After public notice and review as required by the Illinois Endangered Species Protection Act, authorization for incidental take of the species listed above was approved by the Department on July 15, 2016 (hereinafter referred to as the original authorization) (Appendix A). That original authorization remains in effect until July 15, 2031.

Mussel relocation within the proposed bridge corridor occurred from August 1, 2016, through October 25, 2016 (Appendix B). During the relocation, two additional State-listed mussel species were found: purple wartyback (*Cyclonaias tuberculata*) and spike (*Elliptio dilatata*). A conservation plan was received by the Department on January 26, 2017, as a request to add the two additional mussel species to the original authorization for incidental take. The Department requested additional information on February 22, 2017, to make the conservation plan complete as prescribed by Ill. Adm. Code 1080.10. That additional information was received by the Department on February 23, 2017.

Public notice of the DOT' request for an amendment to the original authorization of incidental take adding purple wartyback and spike was published in the Breeze Courier (official State newspaper) on February 28, 2017, and in The Quad-Cities Times on February 28, March 7, and 14, 2017. A copy of the conservation plan was deposited at the Moline Public Library, where it was available for public review. No comments were received from the public.

The Compliance section of the original authorization, except the scientific information under item #4, remains relevant to this amendment and shall apply to the added species. Biological information on the two additional potentially affected species follows:

The **purple wartyback** is an Illinois State-threatened mussel species. It is known to inhabit larger rivers in areas with moderate current and gravel substrates. Purple wartybacks are reported to live up to 20 years, and reach sexual maturity around age 6.

Purple wartybacks are tachytictic, or short-term brooders. Females retain glochidia in their gills from May through August. Glochidial host fish for purple wartybacks include channel catfish (*Ictalurus punctatus*), yellow bullhead (*Ameiurus natalis*), flathead catfish (*Pylodictis olivaris*), and black bullhead (*Ameiurus melas*).

Purple wartyback populations have declined due to habitat loss and degradation. Dams, channelization, and dredging cause increased siltation, physically alter habitat conditions, and block the movement of fish hosts. Purple wartybacks are also susceptible to infestations of invasive zebra mussels, which causes the death of native mussels by attaching to their shells in large numbers and suffocating them.

In Illinois, purple wartybacks have been found in the Mississippi, Kankakee, Vermilion, Fox, and Rock Rivers. They have been found in 11 of 102 Illinois counties. There are currently 39 Element Occurrence Records for purple wartyback in the Illinois Natural Heritage Database. The Department has 11 pending or previously issued Incidental Take Authorizations for purple wartyback in Illinois. Types of projects included dredging, bridge removal and construction/replacement, railroad construction, oil pipeline construction, and diffuser installation. Previous applicants have been required to relocate mussels to suitable habitat prior to the start of construction, minimize instream work, conduct instream work outside of mussel breeding seasons, and enact siltation control measures. This is the first incidental take authorization granted for purple wartyback in Rock Island County.

During the mussel relocation conducted August 1, 2016, through October 25, 2016, two additional mussel species were found, which were not included in the original authorization. One purple wartyback individual was relocated from the new bridge action area. The DOT estimate the potential take of 202 purple wartybacks as a result of construction and demolition.

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The **spike mussel** is an Illinois State-threatened mussel species. It is typically found in small to large rivers, usually occupying sand and gravel substrates. Spike mussels are also occasionally found in reservoirs and lakes, usually associated with outlet habitats dominated by swift currents.

Spike mussels are tachytictic, or short-term brooders. Females brood their young in their gills from May through August before releasing glochidia. Glochidial host fish for the spike mussel include gizzard shad (*Dorosoma cepedianum*), flathead catfish, white crappie, black crappie (*Pomoxis nigromaculatus*), and yellow perch (*Perca flavescens*).

Spike populations have declined due to widespread degradation of habitat throughout its range, caused by hydrologic alteration of streams and watersheds, pollution, and increased sedimentation. Dams, channelization, and dredging can also impact glochidial host fish. Spike mussels are vulnerable to impacts caused by the invasive zebra mussel.

Spike mussels have been found throughout Illinois, including the Mississippi, Illinois, Kaskaskia, Kankakee, Fox, Sangamon, Wabash, and Little Wabash Rivers, in 19 of 102 Illinois counties. There are currently 43 extant Element Occurrence Records for spike mussel in the Illinois Natural Heritage Database. There have been 8 previously issued or currently pending Incidental Take Authorizations for spike mussel in Illinois. Previous projects include bridge replacements, pipeline installation, water treatment outfall, and diffuser installation. Previous applicants have been required to relocate mussels to suitable habitat prior to the start of construction, minimize instream work, conduct instream work outside of mussel breeding seasons, and enact siltation control measures. This is the first authorization to be granted for the take of spike mussels in Rock Island County.

During the mussel relocation conducted August 1, 2016, through October 25, 2016, two additional mussel species were found, which were not included in the original authorization. Two spike mussel individuals were relocated from the new bridge action area. The DOT estimate the potential take of 202 spike mussels as a result of construction and demolition.

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Based on the amount of habitat impacted by this project, the number of known occurrences of the purple wartyback and spike mussels in Illinois, an assessment of the potential effect of this project on individual mussels in the project footprint, the conservation measures included in this authorization for incidental take, and the understanding that vulnerability and recovery information on the species remains limited; the Department has concluded that the taking proposed herein will not reduce the likelihood of survival or recovery of the species in the wild within the State of Illinois, the biotic community of which the species are a part, or the habitat essential to the species' existence in Illinois.

#### **Amendment Terms and Conditions:**

All terms and conditions included in the original authorization and the Conservation Plan submitted by the DOT to the Department are incorporated into this agreement by reference and are made a part thereof.

1. This amendment modifies certain terms and conditions of the original authorization issued to the DOT on July 15, 2015, [IDNR ITA File #56]. This amendment is effective upon the signature of the Department. This amendment and the original authorization may be revoked if the Department finds that the DOT have failed to comply with any of these terms and conditions or have been responsible for the taking of purple wartyback or spike mussels beyond that which is incidental to activities associated with the I-74 Bridge replacement between Moline, Illinois, in Rock Island County, and Bettendorf, Iowa, in Scott County.
2. Upon approval of this amendment by the Department, the terms and conditions of this amendment shall coincide with the original authorization and shall remain in effect through July 15, 2031. All DOT surveying and reporting obligations at the project location shall remain in effect until completed. The Department has determined that minimization, monitoring, and mitigation in the original authorization will bring conservation benefit to the purple wartyback and spike mussels, therefore no additional assurances are required in this amendment.
3. The effective period of this amendment may be altered by mutual written agreement between the DOT and the Department. The Illinois Endangered Species Protection Board shall be notified of any such alteration. Any substantive changes, including but not limited to re-initiation of construction at the project site, a change in the project footprint, or a change in the Illinois endangered or threatened species which could potentially be affected, will require that a new conservation plan be submitted to the Department to initiate the review and public notice process as required by the Act.
4. The original authorization and this amendment are non-transferable.
5. All reports and other documentation required by the original authorization and this amendment shall be submitted to:


Illinois Department of Natural Resources  
Endangered Species Program – Incidental Take Authorization Coordinator  
One Natural Resources Way  
Springfield, IL 62702-1271  
(217)557-8243  
DNR.ITAcoordinator@illinois.gov

The Department's Endangered Species Program shall provide copies of all reports required under the original authorization and this amendment to the Illinois Endangered Species Protection Board and to the Department's Natural Heritage Database.



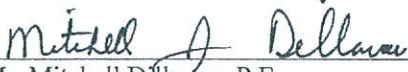
6. The DOT officials identified below are authorized to execute this agreement. Execution by the DOT indicates acceptance of all terms and conditions described in the original authorization and this amendment.
7. The execution of this agreement does not waive or excuse the responsibilities of the DOT to comply with other Federal, State or local regulations, including but not limited to obtaining any required permits for the execution of this project.

*For the Illinois Department of Natural Resources:*

  
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Christopher L. Young, Director  
Office of Resource Conservation

7/20/17  
Date

*For Iowa Department of Transportation:*

  
\_\_\_\_\_  
Mr. Mitchell Dillavou, P.E.  
Highway Division Director

7/11/17  
Date

*For the Illinois Department of Transportation:*

  
\_\_\_\_\_  
Mr. Kevin Marchek, P.E.  
Region 2 Engineer

7-12-2017  
Date



# Illinois Department of Natural Resources

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## **Authorization for Incidental Take and Implementing Agreement**

Pursuant to the Illinois Endangered Species Protection Act (Act) (520 ILCS 10/5.5) and the regulations adopted to implement the Act (17 Ill. Adm. Code 1080), authorization is hereby granted to the Illinois Department of Transportation and the Iowa Department of Transportation (hereinafter referred to collectively as DOT) for the incidental take of Higgins eye (*Lampsilis higginsii*), spectaclecase (*Cumberlandia monodonta*), sheepnose (*Plethobasus cyphus*), butterfly (*Ellipsaria lineolata*), ebonyshell (*Fusconaia ebena*), and black sandshell mussels (*Ligumia recta*). The Illinois Department of Natural Resources (hereinafter referred to as Department) has determined that the taking is incidental to activities associated with the I-74 Bridge replacement between Moline, Illinois, in Rock Island County, and Bettendorf, Iowa, in Scott County. The project area is located in Pool 15 near River Mile 486. The project area is also located within the Mississippi River – Moline Illinois Natural Areas Inventory Site (INAI #1295) and lies directly upstream of Sylvan Slough, an area known for high mussel diversity and abundance.

### **Procedural History**

The Department received a Biological Assessment (BA) containing a Conservation Plan specifically addressing the State of Illinois' requirements (Appendix A of BA) prepared by Stantec Consulting Services for the DOT on May 5, 2016, as a request for authorization for the incidental take of Higgins eye, spectaclecase, sheepnose, butterfly, ebonyshell, and black sandshell mussels. The Department requested additional information on May 11, 2016, to make the conservation plan complete as prescribed by Ill. Adm. Code 1080.10. That additional information was received by the Department on May 13, 2016. The public notice period will be detailed under #6 of the Compliance section below.

### **Compliance with the Illinois Endangered Species Protection Act**

The Act includes six criteria that must be satisfied for the authorization of incidental take of an endangered or threatened species. These criteria and the Department's determination for each are listed below.

1. The taking will not be the purpose of, but will only be incidental to, the carrying out of an otherwise lawful activity:

The stated and apparent purpose of this proposed action is the replacement of the I-74 Bridge across the Mississippi River between Rock Island County, Illinois and Scott County, Iowa to improve capacity, travel reliability, and public safety. The project will include the construction of a new basket handle twin arch bridge, the removal of the existing I-74 suspension bridge, the construction of two storm

sewer outfalls to the Mississippi River, and dredging to allow barge access to staging, construction, and demolition areas. In total (Illinois and Iowa), the new bridge will be 3,372 feet in length and consist of 14 concrete piers supporting the deck. Mussel relocation efforts will begin during the summer of 2016. The construction of the new bridge is scheduled to begin in September 2017 and be completed in November 2020. The demolition of the existing bridge is scheduled to take place from November 2020 to fall 2021. Due to the size of the project and the uncertainty of river conditions, it is understood that the project schedule may be altered. Construction and/or demolition activities will occur year-round. Specifically, temporary impact to suitable mussel habitat at the new bridge could include approximately 6.2 acres of dredging. Permanent impact to suitable mussel habitat at the new bridge includes approximately 0.69 acre due to the placement of piers. The storm sewer outfall projects could temporarily impact 0.028 acre of suitable mussel habitat due to coffer dam installation. In general, the temporary impact action area includes the construction and demolition footprints as well as a 50-foot buffer on both the upstream and downstream sides of the existing and proposed bridges within the Mississippi River. Though construction and demolition activities will occur within and adjacent to the navigation channel, the channel is regularly dredged by the U.S. Army Corps of Engineers and no impacts to mussels are expected; therefore, the navigation channel is not included in the action area. Take of Higgins eye, spectaclecase, sheepsnose, butterfly, ebonyshell, and black sandshell mussels could occur as a result of crushing or burial of individuals left behind following the relocation efforts when construction materials are placed into the riverbed or during dredging. Noise and vibration resulting from coffer dam installation, drilling, and construction may have an adverse effect on some life history stage of the mussels. Some mortality from relocation is expected. Construction and relocation activities may indirectly result in the short-term decrease to reproduction due to stress and/or disturbance to mussels. Construction may cause the displacement of glochidial fish hosts and increase sedimentation in mussel habitat areas. The take of Higgins eye, spectaclecase, sheepsnose, butterfly, ebonyshell, and black sandshell mussels that could result from this project is not the purpose of the DOT's activities, but is incidental to the carrying out of an otherwise lawful activity.

2. The parties to the conservation plan will, to the maximum extent practicable, minimize and mitigate the impact caused by the taking:

Proposed minimization measures were included in the DOT conservation plan.

To meet the "maximum extent practicable" standard, additional minimization and/or mitigation measures may be required beyond those proposed by the DOT, based on the life history needs of the Higgins eye, spectaclecase, sheepsnose, butterfly, ebonyshell, and black sandshell mussels. **All required minimization and mitigation measures are presented under the Authorization section below.**

3. The parties to the conservation plan will ensure that adequate funding for the conservation plan will be provided:

The DOT states that all proposed mitigation will be completed as part of, and not separate from, the construction of the project and in many cases will also be conditions of other permits. Therefore, funding for the mitigation will be included in funding for the overall project. The DOT, along with the Federal Highway Administration, commit to funding construction of the project, and by extension, funding of the mitigation.

4. Based on the best available scientific data, the Department has determined that the taking will not reduce the likelihood of survival or recovery of the endangered species or threatened species in the wild within the State of Illinois, the biotic community of which the species is a part, or the habitat essential to the species' existence in Illinois:

The **Higgins eye mussel** is a Federally-endangered and Illinois State-endangered mussel species. It is a medium-sized mussel that is known to inhabit large rivers. Literature supports that the species' preferred habitat consists of plant-free, stable areas of mixed sand and gravel, and that it is unlikely to be found in areas of shifting sand or silt.

Higgins eye mussels are bradyctictic, or long-term brooders, meaning that the females will retain developing glochidia in their gills over winter and release them the following spring or summer. Higgins eye females will retain glochidia from September through May, with glochidial release occurring from May to August. As with other mussels, Higgins eye glochidia parasitize certain fish species until they grow into juvenile mussels. Possible glochidial host fish species for the Higgins eye include sauger (*Sander canadensis*), largemouth bass (*Micropterus salmoides*), smallmouth bass (*Micropterus dolomieu*), walleye (*Sander vitreus*), yellow perch (*Perca flavescens*), black crappie (*Pomoxis nigromaculatus*), and green sunfish (*Lepomis cyanellus*).

Higgins eye mussels are rare or extirpated from much of their historical range. Their decline is due to poor habitat conditions resulting from human river management, non-point and point-source water and sediment pollution, and the infestation of invasive zebra mussels (*Dreissena polymorpha*). The Higgins eye mussel was the first freshwater mussel species to gain federal protection in the United States. In Illinois, Higgins eye mussels have been found in the Mississippi and Rock Rivers. There are currently nine (9) extant Element Occurrence Records for Higgins eye mussels in the Illinois Natural Heritage Database in four (4) of 102 Illinois counties. The Department has three (3) pending or issued Incidental Take Authorizations for the Higgins eye mussel. Types of projects included dredging, bridge removal and replacement, and pier removal.

In August and September 2014, personnel from Ecological Specialists, Inc., performed surveys for freshwater mussels in and near the area that will be affected by this project. Based on the surveys, an estimated average density of 1,121,871 freshwater mussels (estimated 72,608 listed mussels) may be located within the entire Action Area (old bridge, new bridge and storm sewer outfalls) on the Illinois side of the river. The DOT estimates that this project has the potential to take approximately 3,530 Higgins eye mussels from the Illinois side of the river.

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The **spectaclecase mussel** is a Federally-endangered and Illinois State-endangered mussel species. It is known to inhabit large rivers with swiftly flowing water. They are usually found among boulders in patches of sand, cobble, or gravel in areas of reduced current.

Spectaclecase mussels are thought to be tachytictic, or short-term brooders, releasing their glochidia from early April to late May. Some researchers believe it is possible that spectaclecase mussels are capable of producing two broods per year, one in the spring or early summer and another in the fall; however, this has not been confirmed. Spectaclecase mussels produce the smallest known glochidia of any North American mussel. Glochidia are released in capsules called conglutinates. A single conglutinate may contain ten to hundreds of thousands of glochidia, and females may release between 50 and 90 conglutinates at a time. Despite this extremely high reproductive potential, evidence shows that spectaclecase glochidia have extremely low survival rates to adulthood. Host fish for spectaclecase glochidia are unknown, but glochidia were found in one instance on bigeye chub (*Hybopsis amblops*) and pealip redhorse (*Moxostoma pisolabrum*).

Spectaclecase mussels live close together in colonies, which makes them particularly vulnerable to infestations of the invasive zebra mussel. In Illinois, spectaclecase mussels have been found in the Mississippi River in Hancock, Henderson, Madison, Mercer, and Rock Island Counties. There are currently three (3) extant Element Occurrence Records for spectaclecase mussels in the Illinois Natural Heritage Database in three (3) of 102 Illinois counties. This is the first Incidental Take Authorization written by the Department for spectaclecase mussel.

In August and September 2014, personnel from Ecological Specialists, Inc., performed surveys for freshwater mussels in and near the area that will be affected by this project. Based on the surveys, an estimated average density of 1,121,871 freshwater mussels (estimated 72,608 listed mussels) may be located within the entire Action Area (old bridge, new bridge and storm sewer outfalls) on the Illinois side of the river. The DOT estimates that this project has the potential to take approximately 408 spectaclecase mussels from the Illinois side of the river. However, the primary location of spectaclecase mussels, Pier K of the old



bridge, will not be removed as a minimization measure; thus drastically reducing the potential for take of this species.

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The **sheepnose mussel** is a Federally-endangered and Illinois State-endangered mussel species. It is known to inhabit medium to large rivers in shallow areas of moderate to swift current. The species usually inhabits gravel or gravel mixed with sand, although it has also been found in areas of mud, cobble, and boulders.

Sheepnose mussels are short-term brooders, with reproduction occurring between May and July. Glochidia are released in conglomerates that mimic food organisms of fish, so that they are eaten and glochidia gain access to host fish. Sauger are the only confirmed host fish of sheepnose glochidia, but lab experiments have been successful with flathead minnow (*Pimephales promelas*), central stoneroller (*Campostoma anomalum*), and brook stickleback (*Culaea inconstans*).

Sheepnose mussels are declining in today's rivers due to their management as navigation canals. Dams, channelization, and dredging increase siltation, physically alter habitat conditions, and block the movement of fish hosts. In Illinois, sheepnose mussels have been found in the Mississippi, Rock, Ohio, Wabash, Kaskaskia, and Kankakee Rivers. There are currently nine (9) extant Element Occurrence Records for sheepnose mussels in the Illinois Natural Heritage Database in 11 of 102 Illinois counties. The Department has five (5) pending or issued Incidental Take Authorizations for sheepnose mussels. Types of projects included an oil pipeline, diffuser installation, railroad construction, and a dredge/pier removal.

In August and September 2014, personnel from Ecological Specialists, Inc., performed surveys for freshwater mussels in and near the area that will be affected by this project. Based on the surveys, an estimated average density of 1,121,871 freshwater mussels (estimated 72,608 listed mussels) may be located within the entire Action Area (old bridge, new bridge and storm sewer outfalls) on the Illinois side of the river. The DOT estimates that this project has the potential to take approximately 866 sheepnose mussels from the Illinois side of the river.

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The **butterfly mussel** is an Illinois State-threatened mussel species. It is known to inhabit large rivers in areas of moderate to swift current, and is usually found in substrates of coarse sand and gravel.

Butterfly mussels are long-term brooders, with females retaining developing glochidia in their gills from August until the following July. Known glochidial host fish for this species are freshwater drum (*Aplodinotus grunniens*), green sunfish, and sauger.

Butterfly mussel populations are shrinking due to a decline in habitat conditions associated with river/water management, impacts from the invasive zebra mussel, and from overharvest caused by the button and pearl industries. In Illinois, butterfly mussels have been found in the Mississippi and Ohio Rivers. There are currently 33 extant Element Occurrence Records for butterfly mussels in the Illinois Natural Heritage Database in 12 of 102 Illinois counties. The Department has ten (10) pending or issued Incidental Take Authorizations for butterfly mussels. Types of projects included dredging, construction of offshore structures, bridge removal and replacement, riprap installation, and boat dock installation.

In August and September 2014, personnel from Ecological Specialists, Inc., performed surveys for freshwater mussels in and near the area that will be affected by this project. Based on the surveys, an estimated average density of 1,121,871 freshwater mussels (estimated 72,608 listed mussels) may be located within the entire Action Area (old bridge, new bridge and storm sewer outfalls) on the Illinois side of the river. The DOT estimates that this project has the potential to take approximately 14,764 butterfly mussels from the Illinois side of the river.

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The **ebonyshell mussel** is an Illinois State-endangered mussel species. It is known to inhabit large rivers in areas of swift current. It is usually found in stable sand or gravel substrate.

Ebonyshell mussels are short-term brooders. Reproduction takes place from May to early fall, after which glochidia are released. The primary host fish for ebonyshell glochidia is the skipjack herring (*Alosa chrysochloris*); although literature supports that it is possible that largemouth bass, white crappie (*Pomoxis annularis*), and black crappie could also be hosts for this species.

Ebonyshell was historically the most abundant mussel species in the Upper Mississippi River, but populations have declined dramatically over the past century. One cause of the ebonyshell's decline was that its pearly-white interior shell was highly prized by button-makers, which led to its overharvest. In Illinois, they have been found in the Mississippi, Illinois, Ohio, Wabash, and Little Wabash Rivers. There are currently 12 extant Element Occurrence Records for ebonyshell mussels in the Illinois Natural Heritage Database in eight (8) of 102 Illinois counties. The Department has issued one (1) previous Incidental Take Authorization for ebonyshell for a dredging project in the Ohio River.

In August and September 2014, personnel from Ecological Specialists, Inc., performed surveys for freshwater mussels in and near the area that will be affected by this project. Based on the surveys, an estimated average density of 1,121,871 freshwater mussels (estimated 72,608 listed mussels) may be located within the entire Action Area (old bridge, new bridge and storm sewer outfalls) on the Illinois side of the river. No live ebonyshell mussels were found in the surveyed areas; however one weathered dead ebonyshell was collected. The DOT

do not estimate any take of ebonyshell mussels from the Illinois side of the river as a result of this project.

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The **black sandshell** mussel is an Illinois State-threatened mussel species. It is found in medium to large rivers in areas with strong currents. The species is known to prefer substrates of coarse sand, gravel, cobble, or silt.

Black sandshell mussels are long-term brooders, with females retaining developing glochidia in their gills from August until the following July, after which glochidia are released. Gravid female black sandshells are known to display their marginal papillae, moving them in a way that attracts fish hosts before releasing the parasitic glochidia. Black sandshell host fish include walleye, bluegill (*Lepomis macrochirus*), largemouth bass, sauger, white crappie, and many others that have been suggested as possible hosts.

Black sandshell populations have declined due to habitat degradation. In Illinois, black sandshells have been found in the Mississippi, Kaskaskia, Vermilion, Ohio, Kankakee, Rock, Iroquois, and Little Wabash Rivers, as well as several smaller creeks and tributaries. There are currently 97 extant Element Occurrence Records for black sandshell mussels in the Illinois Natural Heritage Database in 30 of 102 Illinois counties. The Department has 23 pending or issued Incidental Take Authorizations for black sandshell. Types of projects included dredging, bridge removal and construction/replacement, boat dock construction, railroad construction, pier removal, dam removal, piling installation, riprap installation, oil pipeline construction, and diffuser installation.

In August and September 2014, personnel from Ecological Specialists, Inc., performed surveys for freshwater mussels in and near the area that will be affected by this project. Based on the surveys, an estimated average density of 1,121,871 freshwater mussels (estimated 72,608 listed mussels) may be located within the entire Action Area (old bridge, new bridge and storm sewer outfalls) on the Illinois side of the river. The DOT estimates that this project has the potential to take approximately 53,312 black sandshell mussels from the Illinois side of the river.

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Based on the amount of habitat impacted by this project, the number of known occurrences of the Higgins eye, spectaclecase, sheepnose, butterfly, ebonyshell, and black sandshell mussels in Illinois, an assessment of the potential effect of this project on individual mussels in the project footprint, and the conservation measures included in this authorization for incidental take; the Department has concluded that the taking proposed herein will not reduce the likelihood of survival or recovery of the Higgins eye, spectaclecase, sheepnose, butterfly, ebonyshell, and black sandshell mussels in the wild within the State of Illinois,

the biotic community of which the species are a part, or the habitat essential to the species' existence in Illinois.

5. Any measures required under Section 5.5(b)(6) of the Act will be performed:

These measures are listed below under "Authorization." This authorization is, by definition, subject to those terms and conditions and the signatures of representatives of the DOT indicate their commitment to performing those measures.

6. The public has received notice of the application and has had the opportunity to comment before the Department made any decision regarding the application:

Public notice of the DOT' request for authorization of incidental take was published in the Breeze Courier (official state newspaper) on May 19, 2016, and in The Quad-Cities Times on May 19, 26, and June 2, 2016. A copy of the conservation plan was deposited at the Moline Public Library, where it was available for public review. The deadline for public comment was July 2, 2016. Four (4) comments were received from the public. The comments were transmitted to DOT on June 13, 2016. An analysis of the comments was received by the Department on July 12, 2016.

### Authorization

It is the determination of the Department that the measures that will be implemented by the DOT will adequately minimize and mitigate the anticipated taking of Higgins eye, spectaclecase, sheepnose, butterfly, ebonyshell, and black sandshell mussels incidental to activities associated with the I-74 Bridge replacement over the Mississippi River between Rock Island County, Illinois, and Scott County, Iowa. Further, the Department has concluded that the take authorized herein will not reduce the likelihood of survival or recovery of the Higgins eye, spectaclecase, sheepnose, butterfly, ebonyshell, and black sandshell mussels in the wild within the State of Illinois, the biotic community of which the species are a part, or the habitat essential to the species' existence in Illinois. Additional listed mussel species are known to inhabit the Mississippi River, this agreement does not authorize take of any species except Higgins eye, spectaclecase, sheepnose, butterfly, ebonyshell, and black sandshell mussels.

All terms and conditions included in the aforementioned BA and Conservation Plan submitted by the DOT to the Department are incorporated into this agreement by reference and are made a part thereof.

Pursuant to Section 5.5 of the Illinois Endangered Species Protection Act [520 ILCS 10/5.5] and the Administrative Rules for the Incidental Taking of Endangered and Threatened Species [Ill. Adm. Code 1080.40(b)], this authorization is issued subject to the following terms and conditions, which may include additions or modifications to the minimization and mitigation measures proposed by the DOT in the conservation plan:

1. This authorization is effective upon the signature of the Department and shall remain in effect for a period of **fifteen (15) years** from the date of the Department signature, unless terminated by written agreement of all parties.

This authorization may be revoked pursuant to the Act and Ill. Adm. Code 1080.80(b) if the Department finds that the DOT has failed to comply with any of these terms and conditions or has been responsible for the taking of Higgins eye, spectaclecase, sheepsnose, butterfly, ebonyshell, or black sandshell mussels beyond that which is incidental to activities associated with the I-74 Bridge replacement over the Mississippi River between Rock Island County, Illinois, and Scott County, Iowa.

2. The effective period of this authorization may be altered by mutual written agreement between the DOT and the Department. The Illinois Endangered Species Protection Board shall be notified of any such alteration.

Any substantive changes, including but not limited to a change in the project footprint or a change in the State-listed species which could potentially be affected, will require that a new conservation plan be submitted to the Department to initiate the review and public notice process as required by the Act.

3. This authorization is non-transferable.
4. On-site personnel shall be educated on the sensitive biological resources in the area, the identification of listed mussel species, regulations protecting the species, where the species might be found, avoidance areas, travel restrictions for equipment and vehicles, how to report sightings or incidents that may involve take, and the importance of avoiding take of the species. The DOT shall submit a copy of all education materials to the Department.
5. The Department reserves the right of entry by its staff or representatives to inspect species, potential habitat, and species management practices.
6. Biological consultants employed by the DOT shall hold the necessary permits for work with non-listed and listed species; these include an Illinois Department of Natural Resources Scientific Collection Permit and an Illinois Department of Natural Resources Endangered Species Permit.
7. The DOT shall notify the Department's Endangered Species Program of construction commencement and completion of the I-74 Bridge replacement project. Any discoveries of additional listed species beyond those identified in this agreement shall be reported to the Department within 48 hours accompanied by location information (photograph and GPS coordinates).



8. The DOT shall conduct, or cause to be conducted, the following pre-construction or construction efforts:
- a. Staging area locations will be chosen according to the following restrictions - on the Illinois side, no construction access within Sylvan Slough and extending upstream of the proposed bridge corridor beyond 50-foot buffer.
  - b. All barges and watercraft used for construction activities shall be inspected for the presence of zebra mussels prior to placing the barges into the Mississippi River and shall be completely out of water for 10 days to ensure proper drying and reduce potential infestation by zebra mussels.
  - c. Workers shall be transported to and from the construction/demolition areas daily via either a small watercraft or work barge.
  - d. Materials shall be transported to and from the construction/demolition areas by work barge as needed.
  - e. Dredged material will not be placed back into the river. Areas disturbed by dredging shall be backfilled with a special revetment.
  - f. Floating silt curtains shall be installed prior to construction of the bridge to retain sediment created by construction. This action protects the City of Moline's drinking water intake structure, in addition to avoiding and/or minimizing the effects of sedimentation on mussels.
    - i. Silt curtains shall be installed.
    - ii. A permitted diver shall be present to remove and relocate any mussels that may be present at proposed silt curtain anchor locations. Mussels shall be handled and relocated according to the protocol described below in Authorization condition # 9.
    - iii. Silt curtains shall be inspected regularly and subject to routine maintenance.
    - iv. Any accumulated debris on the river bottom and at the surface caught by the silt curtain shall be removed before curtain removal.
  - g. Debris shall not be allowed to collect at the bottom of the river. Any debris that falls into the water shall be removed by the contractor during the same work day as soon as is practicable.
  - h. All spoil from construction shall be placed on barges and taken offsite. No fill material shall be left in the river.
  - i. The removal of the existing I-74 Bridge shall include the removal of the bridge deck and all existing piers with the exception of Pier K in Sylvan Slough. This pier shall remain intact to minimize effects on the mussel bed at that location. The contractor shall be restricted from impacting the river bottom within a 16.4-foot buffer of Pier K.
  - j. No explosive demolition or dropping of materials into the river shall take place on the Illinois side of the river.
  - k. No causeway or elevated rock work platform shall be installed in the river.
  - l. Any and all other construction provisions and work zone restrictions as identified in the BA shall apply, including but not limited to those provisions outlined in Appendix C (Iowa DOT Special Provisions).

9. The DOT shall conduct, or cause to be conducted, the following mussel survey and relocation efforts:
- a. A thorough search for freshwater mussels within the footprint of the construction of the replacement bridge. Areas to be searched shall include the projected locations for the first five (5) piers on the Illinois bank and within a 10-meter buffer of each of those five piers (each pier/buffer area will be approximately 90 meters long by 30 meters wide). Due to the number of mussels to be relocated (estimated 268,212 listed and non-listed individuals), the relocation effort is anticipated to take approximately 60 days. Therefore, mussels will be relocated from the removal areas between July and September 2016 (the year prior to initiation of construction). The search shall be conducted during biologically suitable mussel relocation periods. All freshwater mussels found shall be identified to species, marked, and enumerated. Those conducting the search must be qualified at accurate identification of freshwater mussel species. All native freshwater mussels found during this search shall have zebra mussels removed from their shells and be relocated to suitable habitat as discussed below under (g).
  - b. A thorough search for freshwater mussels within the footprint of the existing bridge prior to demolition and removal. Areas to be searched shall include the area underneath the bridge where debris could fall. This search shall be conducted prior to initiation of demolition of the old bridge and during biologically suitable mussel relocation periods. All freshwater mussels found shall be identified to species, marked, and enumerated. Those conducting the search must be qualified at accurate identification of freshwater mussel species. All native freshwater mussels found during this search shall have zebra mussels removed from their shells and be relocated to suitable habitat as discussed below under (g).
  - c. All mussels shall be salvaged from any cofferdam areas.
  - d. Relocation sites have been pre-approved by the Department. In order to accommodate the large-scale relocation effort prior to construction, three sites were identified by Ecological Specialists, Inc. as potential suitable relocation areas for all mussel species. In addition, two relocation sites were identified specifically due to their suitability for the spectaclecase mussel. This species is a habitat specialist and primarily occurs on substrate composed of large rock. Surveys delineated the existing mussel bed at each location and estimated species richness and density of mussels at each location to determine suitability as relocation areas.
  - e. Mussels will not be relocated when air temperatures are at or below 32 degrees Fahrenheit, nor when water temperatures are at or below 40 degrees Fahrenheit. All mussels will be held in mesh bags suspended in the river or in containers of water changed every hour (every half-hour when air temperatures are at or above 80 degrees Fahrenheit). Water in containers shall be taken from the river where the mussels were collected. No mussels shall be held for more than three (3) hours before being returned to the locality from which they were taken or previously authorized relocation sites.

- f. Any and all other mussel survey and relocation provisions as identified in the BA shall apply, including but not limited to those provisions outlined in Appendix D (Mussel Relocation Plan).
  - g. **A report including, but not limited to, the survey methodology utilized, the species and numbers of mussels located, the age and size of each individual sampled, and maps of the area searched and the relocation site shall be provided to the Department within 90 days of completion of the survey and relocation effort.**
10. The DOT shall conduct, or cause to be conducted, the following post-construction freshwater mussel monitoring efforts:
- a. Monitoring shall be conducted within and adjacent to the footprint of the new I-74 Bridge. The objective will be to examine the effects of construction on the mussel community and the rate of recolonization. Baseline data related to mussel density and species composition shall be collected prior to the start of construction. Monitoring shall occur during Year 1 or the first safe time after pier installation and in the 3<sup>rd</sup>, 6<sup>th</sup>, and 9<sup>th</sup> years following construction completion.
  - b. Recipient site monitoring of the three (3) species listed both federally and by the State shall take place in select relocation areas. Marked specimens of Higgin's eye, sheepnose, and spectaclecase mussels will be placed within cells in a 5x5 grid. Each specimen shall be measured, aged, sexed, and marked with a unique identification number. Spectaclecase mussels will also be marked with a PIT tag. The grid cell in which each individual is placed shall be recorded. Monitoring shall then be conducted in order to relocate the marked individuals and collect data regarding their survival, movement, and growth over time. Monitoring shall take place annually for the first two years, along with the 4<sup>th</sup>, 7<sup>th</sup>, and 10<sup>th</sup> years following mussel relocation.
  - c. Mussel surveys shall not take place when air temperatures are at or below 32 degrees Fahrenheit, nor when water temperatures are at or below 40 degrees Fahrenheit. If mussels are removed from the water during surveys, they will be held in mesh bags suspended in the river or in containers of water changed every hour (every half-hour when air temperatures are at or above 80 degrees Fahrenheit). Water in containers shall be taken from the river where the mussels were collected. No mussels shall be held for more than three (3) hours before being returned to the locality from which they were taken or previously authorized relocation sites.
  - d. Any and all other mussel monitoring provisions as identified in the BA shall apply, including but not limited to those provisions outlined in Appendix G (Monitoring Plan).
  - e. **Detailed reports including, but not limited to, the survey methodology utilized, the species and numbers of mussels located (noting any marked individuals), the age and size of each individual sampled, and a map of the species locations, shall be provided to the Department within 90 days of completion of each monitoring event.**
11. The DOT shall mitigate for the potential taking of listed species to the maximum extent practicable by bringing conservation benefit to the species potentially impacted.

On May 10, 2016, an Intergovernmental Agreement (IGA) was executed between the Department, the Iowa Department of Natural Resources, and the USFWS regarding the collaborative development of a conservation strategy to serve as mitigation for take associated with the I-74 Bridge project. As a result of this collaboration, the following mitigation proposals were agreed to by the DOT for implementation:

- a. A large-scale study of Pool 15 will occur in three phases to map habitat and mussel distribution consisting of compilation and mapping of existing data, quantitative sampling to map the distribution of existing mussel beds in channel border habitat and provide calibration for larger scale sampling, poolwide sampling to determine density and population estimates. Data from this study will help resource agencies track impacts from the I-74 Bridge project on mussel resources and guide future conservation efforts in Pool 15.
- b. A study will be conducted to investigate the effects of increasing resident mussel density at varying rates resulting from the I-74 Bridge project mussel relocation. A subset of relocated mussels will be placed at varying densities within the three general (not spectaclecase) relocation sites. The sites will be monitored to determine whether different densities persist or if the beds return to pre-relocation numbers. Monitoring shall occur annually for the first two years and in the 4th, 7th, and 10th years following mussel relocation. Data from this study will provide valuable information on the potential carrying capacity of mussel beds and inform future relocation efforts.
- c. A two-year mussel education and outreach staff position to serve as the point of contact, to develop education materials, to conduct classroom and public interpretive outreach, to perform media and community education, and to develop and coordinate a social media presence. The staff will educate on both the ecology of mussels and bridge demolition/construction techniques. A document may be developed through this position to address best management practices for future bridge projects that have the potential to impact mussel resources.
- d. A five-year effort to inoculate host fish with mussel glochidia and perform free release of 10,000 inoculated fish annually near the project impact in cooperation with the Genoa National Fish Hatchery's Native Mussel Recovery Program. This effort will assist with repopulation of impacted areas and offset the impacts from bridge construction and demolition. The resource agencies will determine which mussel species and host fish species will be stocked based on the data collected from the mussel relocation and the impacts to mussel beds within the action area.

Above synopses are abbreviated descriptions of the agreed-to mitigation projects. Implementation of each of the mitigation projects shall follow the more detailed proposals provided to the DOT with further negotiation, as necessary, with the natural resource agencies. Mitigation value is estimated to be in excess of \$545,000.00.

Mitigation valuations are based on the Department's best current understanding of the species life history needs, real estate values, and impact analysis relevant to the project site's proposed conceptual design elements available at the time of review.

12. As stated previously in this agreement and to emphasize that it relates not only to the project's construction schedule but also to monitoring, relocation, and mitigation studies; due to the size of the project and the uncertainty of river conditions, it is understood by the Department that the schedule may be altered.

13. The DOT shall submit reports on all surveys within 90 days of survey completion.

14. All reports, notifications, and other project documentation shall be submitted to:

Illinois Department of Natural Resources  
Office of Resource Conservation  
Endangered Species Program – Incidental Take Authorization Coordinator  
One Natural Resource Way  
Springfield, IL 62702-1271

(217)557-8243

[DNR.ITAcordinator@illinois.gov](mailto:DNR.ITAcordinator@illinois.gov)

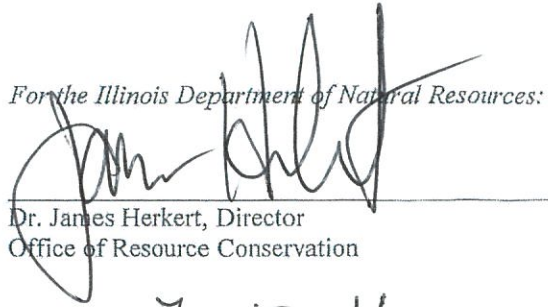
The Department's Endangered Species Program shall provide all reports required under this agreement to the Illinois Endangered Species Protection Board and to the Department's Natural Heritage Database.

15. The DOT officials identified below are authorized to execute this agreement. Execution by the DOT indicates acceptance of all terms and conditions described in this authorization.

16. The execution of this agreement does not waive or excuse the responsibilities of the DOT to comply with other Federal, State, or local regulations, including but not limited to obtaining any required permits for the execution of this project.



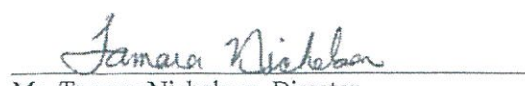
*For the Illinois Department of Natural Resources:*

  
\_\_\_\_\_  
Dr. James Herkert, Director  
Office of Resource Conservation

Date

7-15-16


*For Iowa Department of Transportation:*

  
\_\_\_\_\_  
Ms. Tammy Nicholson, Director  
Office of Location and Environment

Date

7/13/2016

*For the Illinois Department of Transportation:*

  
\_\_\_\_\_  
Mr. Kevin Marchek, P.E.  
Region 2 Engineer

Date

7-13-16

**Unionid Relocation and Baseline Data  
Collection for Unionid Monitoring Studies,  
Interstate 74 Bridge Replacement Project,  
Mississippi River Pool 15**

**Prepared for:**

**Stantec Consulting Services, Inc.**  
Independence, Iowa

**Prepared by:**

**Ecological Specialists, Inc.**  
O'Fallon, Missouri  
An EcoAnalysts Company

**February 2017**

(ESI Project No. 16-006)

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Mr. Robert Williams served as ESI's dive coordinator, and Mr. Kendall Cranney and Mr. Chet Clark served as additional dive supervisors. Mr. Austin Barrett, Mr. Justin Brewster, Mr. Chris Brouillette, Mr. Trevor Davenport, Mr. Benjamin Dunn, Mr. Jeremy Gottman, Mr. Chad Gray, Mr. Jerry Holmes, Mr. Nikolaus Liverar, Mr. Jacob McCarthy, and Mr. Levi Williams (all of ESI) and Mr. Mark Eubank, Mr. Hayden Aschim, Mr. John Crossen, Mr. Dave Henry, Mr. Jacob Hula, Mr. Jason Klett, Mr. Chase Morris, Mr. Patrick Robertson, Mr. Robert Samson, Mr. Wes Steinke, Mr. Benjamin Vinz, Mr. Thurlow Weed, and Mr. Chris West (all of GOE International, LLC) served as divers.

Mr. Luke Hodges, Mr. Ryan Rasmussen, Ms. Lisa Davila, Ms. Hannah Helms, Mr. Rick Sanderson, Ms. Katie Whitehead, Ms. Nissa Wilson, Mr. Jon Currie, Mr. Cody Fleece, Ms. Kim Grmusich, Mr. Matt Knicklebine, Mr. Aaron Kwolek, Mr. Dillon McNulty, Ms. Abby Medis, Mr. Jeff Metcalf, Mr. Josh Otten, Mr. Gabe Pelletier, Ms. Giselle Rogers, Mr. Dan Schepis, Ms. Angela Sjollema, Ms. Kari Soltau, Ms. Molly Stephenson, and Ms. Hannah Stoffs (all of Stantec) assisted with marking, sorting, and processing unionids and recording data.

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Mr. Ford and Ms. Dunn (ESI) assisted with data QA/QC and report preparation.

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## 1.0 Introduction

The Iowa and Illinois Departments of Transportation are replacing the Interstate 74 bridge over the Mississippi River. The existing bridge is located in Pool 15 near river mile 486, and connects the cities of Bettendorf, Iowa (Scott County) and Moline, Illinois (Rock Island County; Figure 1-1). Pool 15 is known to harbor a species-rich unionid (freshwater mussel) community, including several federally endangered species, and the bridge project area overlaps the Sylvan Slough *Lampsilis higginsii* Essential Habitat Area (EHA). Therefore, Ecological Specialists, Inc. (ESI) was contracted to conduct a mussel survey in the bridge project area in 2014. The 2014 survey results indicated that a dense, species-rich mussel bed was present on the Illinois (left descending) bank beneath both the proposed and existing bridge alignments (Table 1-1; ESI, 2014). Three (3) federally endangered species (*Cumberlandia monodonta*, *Plethobasus cyphus*, *L. higginsii*) and 2 Illinois threatened species (*Ellipsaria lineolata*, *Ligumia recta*) were collected within this bed. Although unionid abundance was lower, *C. monodonta*, *L. higginsii*, and the Iowa threatened *E. lineolata* were collected near the existing bridge on the Iowa bank as well (Table 1-1; ESI, 2014).

A mussel mitigation task force was formed following the 2014 survey to aid in developing mitigation options for the bridge project. This task force was composed of representatives from ESI, Stantec Consulting Services, Inc. (Stantec), Iowa Department of Transportation, Illinois Department of Transportation, Iowa Department of Natural Resources (IADNR), Illinois Department of Natural Resources (ILDNR), U.S. Fish and Wildlife Service (USFWS), Illinois Natural History Survey, and the Federal Highway Administration. The task force determined that unionids would need to be relocated from project impact areas, and proposed additional studies and monitoring projects to mitigate impacts to unionids. A Biological Assessment (BA) was prepared to evaluate effects of the bridge replacement project on federally endangered species (Stantec, 2016). A Conservation Plan (CP) was prepared to evaluate effects of the bridge replacement project on Illinois state threatened and endangered (T&E) species (Appendix A of the BA). The BA identified the Action Area for the bridge project; the Action Area for the new bridge includes the construction footprint for the new bridge plus a 15 m (50 ft) buffer on the upstream and downstream sides (Figure 1-1). The Action Area extends from the shore to the navigation channel on both banks. Because it is regularly dredged by the U.S. Army Corps of Engineers and no impacts to unionids are expected, the navigation channel is not included in the Action Area. Also included in the Action Area is a small, approximately 15 m (50-ft) square area on the Illinois bank downstream of the new bridge corridor that may be impacted by construction of a stormwater outfall.

USFWS issued a final Biological Opinion (BO) in response to the BA on July 14, 2016. The BO concluded that the bridge project would not jeopardize the continued existence of *C. monodonta*, *P. cyphus*, or *L. higginsii*, but that incidental take of these species would occur. Therefore, several Reasonable and Prudent Measures (RPMs) were identified to minimize impacts to federally listed species. Several RPMs pertained to unionid relocation and monitoring, including:

- Relocation of unionids from construction and demolition areas to approved relocation areas,
- Sampling within mussel relocation areas before and after relocation to assess the health of relocated unionids, and

- Sampling within the new bridge Action Area before and after bridge construction to evaluate impacts of construction on unionids.

An Incidental Take Authorization (ITA) was also issued in response to the CP to allow the incidental take of Illinois state T&E species. ILDNR, Iowa DOT, and Illinois DOT signed the Incidental Take Authorization in July 2016. The RPMs were included in the ITA as mitigation. ESI was contracted to conduct baseline (pre-relocation) sampling in the new bridge Action Area and relocation areas, and to relocate unionids from direct impact areas in the new bridge Action Area. Baseline data will be compared to data collected in future years to evaluate impacts of construction, unionid recolonization within the construction area, and survival and health of relocated unionids.

## 2.0 Methods

### 2.1 Construction area baseline sampling

The RPMs set forth in the BO and ITA included sampling in the new bridge footprint to quantify the effects of construction on unionids and to determine the rate at which unionids recolonize the construction area. The new bridge Action Area was sampled prior to relocating unionids to provide baseline (pre-relocation and pre-construction) data on the unionid community. Quantitative samples were used to determine unionid density and community metrics. To achieve a confidence interval within 20-25% of the mean, 100 samples were collected in the Illinois portion of the Action Area. Samples were arranged in a three random start design (Strayer and Smith, 2003) for statistical validity. Due to high variability in the Iowa Action Area, and because depth is great enough that construction barges should not directly impact the substrate, samples in the Iowa Action Area were concentrated around the shoreward pier, where unionids were most abundant in the 2014 survey. Twenty (20) randomly distributed quantitative samples were collected in and adjacent to the pier footprint, and 30 randomly distributed samples were collected in the area between the shoreward 2 piers. For each sample, a diver excavated all substrate within a 0.25-m<sup>2</sup> quadrat frame into an attached mesh bag (6 mm mesh). Substrate was sieved through 12 and 6 mm sieves and all unionids were retrieved from the sample. Unionids were identified to species, measured (length in mm), and aged (external annuli count). Height and width (mm) were also measured for federally endangered species, and *L. higginsii* were marked with a unique ID number using a Dremel tool. Zebra mussel infestation was recorded for all live unionids. Dead shells were identified and categorized as either fresh dead (dead within the past year, nacre shiny, hinge flexible, valves attached, with or without tissue), weathered dead (dead many months to years, nacre chalky, hinge brittle, valves typically separated, periostracum intact), or subfossil (dead many years to decades, periostracum eroded, valves separate, very chalky). Fresh dead shells were counted to estimate mortality. At least 1 individual of each species was photographed, and a dead shell of each species (if available) was retained as a voucher. Depth, substrate composition, and GPS coordinates were recorded for each sample. Density estimates in 2014 and 2016 were compared using SYSTAT 13 (Systat Software, San Jose, CA). Data were first tested for normality. Pairwise comparisons between years were then made using the Wilcoxon signed-rank test.

### 2.2 Relocation

#### *Illinois bank*

Eleven (11) piers will be constructed on the Illinois bank (Figure 2-1). Unionid density was high in the shoreward portion of the area, averaging 31.9 mussels/m<sup>2</sup> from the Illinois bank to halfway between Piers 5 and 6. Density decreased sharply to 0.4 mussels/m<sup>2</sup> riverward of this point (ESI, 2014). Much of the area from the bank to Pier 5 is <2 m (6 ft) deep (based on bathymetry data from the U.S. Army Corps of Engineers) and may need to be dredged to allow barge access. Although both pier construction and dredging may affect unionids, to reduce the relocation effort to a more manageable level, unionids were only relocated from the first 5 piers on the Illinois bank, plus 10-m (33-ft) buffers around each pier footprint. Unionids were also relocated from the outfall construction area downstream of the new bridge corridor (Figure 2-1).

Each pier plus buffer area was approximately 90 m (295 ft) long and 30 m (98 ft) wide. A 30 m (98 ft) x 91 m (300 ft)

grid was established at each pier using lines, weights, and buoys. Each grid was then divided into 60 3 m (10 ft) x 15 m (50 ft) cells (Figure 2-2). An additional row of cells (3 m [10 ft]) was added to the upstream and downstream ends of the Pier 1 search grid, as this pier was slightly longer than the others. A diver searched each cell by hand, disturbing the top 10 cm (4 inches) of substrate, and collecting all unionids encountered. The number of unionids collected in each cell was recorded. To ensure that at least 90% of unionids were removed, additional passes of each cell were conducted until the final pass yielded <10% of the cumulative number of unionids collected in that cell. Different divers were used for each pass as an additional quality assurance measure. On average, approximately 55-60 dive days were spent in each pier. The outfall area was also delineated by lines, weights, and buoys, but was not gridded off into smaller cells. This area was searched to diminishing returns in the same manner as the pier areas. Approximately 2 dive days were spent in the outfall area. A dive day consisted of 6-8 hours of diving with one diver in the water.

Collected mussels were brought to the surface, counted, and placed in mesh bags suspended in a holding tank equipped with a well pump to refresh the water or in buckets before being transported to the processing area by boat. The processing area was located on the Illinois bank approximately 100 m (330 ft) downstream of the existing bridge. Mussels were briefly exposed to air during transport, but were held in water (holding tanks or buckets) while awaiting processing. Buckets were kept in the shade on the boats and banks when air temperatures were high, and water was changed frequently to ensure that the temperature remained similar to the ambient river temperature and that dissolved oxygen was not depleted. All unionids were first marked with a slash on the anterior margin of the shell using a Dremel tool, with the exception of *C. monodonta* and individuals too small to etch without damaging the shell. Common and state-listed species were distributed among relocation areas at 3 unionid beds identified in 2015 (Illiniwek Park, Eagle's Landing, and Upstream; Figure 2-3). Two (2) relocation areas were established in each bed, resulting in 6 possible areas in which unionids could be placed. Individuals were assigned to 1 of the 6 relocation areas using a random number list to avoid bias. Following sorting, unionid data were recorded; unionids were processed in groups based on their assigned relocation area. Common species were identified and categorized as adult (>5 years old) or juvenile (≤5 years old). State T&E species were identified, measured (length in mm), and aged (external annuli count); sex was determined for sexually dimorphic species, and females were checked for gravidity. For consistency, this information was recorded for species listed as T&Es in either or both Illinois and/or Iowa. Processed unionids were placed in mesh bags labeled with their designated relocation area and were held in flowing water or holding tanks with well pumps prior to being transported to the relocation areas by boat. Unionids were distributed throughout their respective relocation area from the surface.

Federally endangered species were handled separately. *Lampsilis higginsii* and *P. cyphus* were measured (length, width, and height in mm), aged, and etched with a unique ID number; sex was also determined for *L. higginsii*, and females were checked for gravidity. Each individual was then randomly assigned to a placement grid. Three (3) 5 m x 5 m (16 ft x 16 ft) grids were established for placement of *L. higginsii*, one in each of the unionid beds (Illiniwek Park, Eagle's Landing, and Upstream; ESI, 2015) used for placement of other species (Figure 2-3). Two (2) 5 m x 5 m (16 ft x 16 ft) grids were established for placement of *P. cyphus*, 1 in the Illiniwek Park bed and 1 in the Eagle's Landing bed. Grid

locations were originally selected by generating random points within the unionid bed boundaries. However, the original *L. higginsii* grids at Eagle's Landing and Upstream fell in areas with substrate that appeared suitable at first, but scoured down to bedrock within the first week of the relocation. These grids were thoroughly searched for any remaining *L. higginsii* that had been placed up until that time, and were then removed and reestablished in a new location. No *P. cyphus* had yet been placed in the grid at Eagle's Landing, so this grid was not searched before removing it. To establish the new grids, an experienced diver searched for a more suitable grid site near relocation areas used for common species placement, as these areas were known to contain suitable habitat. Both the *L. higginsii* and *P. cyphus* grids at Eagle's Landing were reestablished in new locations, as was the *L. higginsii* grid at Upstream. Each grid was divided into 4 cells. Individuals were placed in each cell such that resident unionid density was not increased by more than 50%; 5 quantitative samples were collected within each grid prior to the relocation to estimate resident density. A random number list was used to assign individuals to a cell (numbered 1 through 4) within their respective grids. The unique ID number and placement location (cell number) of each individual was recorded to facilitate future monitoring.

*Cumberlandia monodonta* were measured (length, width, and height in mm) and aged, and a PIT tag was affixed to the dorsal margin of the shell. Monitoring of previously relocated *C. monodonta* (ESI, 2016) suggested that the Sylvan Slough site, located just downstream of the Rodman Avenue bridge, was most suitable for placement of additional individuals; therefore, all *C. monodonta* encountered in this relocation were placed at the Sylvan Slough site.

*Cumberlandia monodonta* were hand-placed in suitable habitat near the previously relocated individuals.

#### *Iowa bank*

Three piers will be constructed on the Iowa bank (Figure 2-4). Mussel density on the Iowa bank averaged 2.1 mussels/m<sup>2</sup>. Substrate in the proposed pier construction areas (determined during the 2014 survey; ESI, 2014) was primarily bedrock or sand, and most mussels were found in small patches of sand, silt, and clay. Additionally, most of the area between the piers is >2.4 m (8 ft) deep and should not be affected by construction equipment. Unionids were therefore relocated from the new pier construction areas plus a 5-m (16-ft) buffer around each proposed pier footprint (Figure 2-4). The 2 shoreward piers plus buffers were approximately 80 m (262 ft) long x 20 m (66 ft) wide, while the riverward pier plus buffer was approximately 100 m (328 ft) long x 40 m (131 ft) wide. Each pier plus buffer area was marked with buoys at the corners. Divers conducted sweeps throughout each pier area, focusing on patches of more suitable habitat and collecting all unionids encountered. Because fewer unionids were expected to occur on the Iowa bank, the Iowa piers were not searched to diminishing returns in the same manner as the Illinois piers. Instead, divers searched each pier area for a fixed amount of time. Approximately 2.5 dive days were spent searching the shoreward 2 piers, and 2 dive days were spent in the riverward pier. A dive day consisted of 6-8 hours of diving with one diver in the water.

Collected mussels were transported to a processing location on the Iowa bank beneath the existing bridge. All unionids were etched with a slash on the anterior edge of the shell using a Dremel tool, with the exception of *C. monodonta* and individuals too small to etch without damaging the shell. Common species were identified and counted as adults or

juveniles, and state-listed species were measured and aged. A denser assemblage of mussels ( $8/m^2$ ) was found within 70 m (230 ft) of the Iowa bank between the new bridge corridor and the casino boat upstream (Figure 2-5). This area, beginning approximately 100 m (330 ft) upstream of the new bridge corridor, was to be used for placement of common and state-listed species. One hundred (100) randomly distributed quantitative samples were collected within the area to estimate baseline unionid density. Samples were collected and unionids were processed using the same methods as the construction area sampling (described above). The casino boat was still present during initial sampling, but was removed from the area during the relocation. As the boat is not expected to return to the area, IADNR suggested placing some common unionids in the casino boat footprint. A brief reconnaissance dive in the casino boat footprint suggested that habitat quality was lower than that within the originally proposed relocation area. Therefore, a few very common and/or tolerant species (*Amblema plicata*, *Lampsilis cardium*, *Leptodea fragilis*, *Potamilus alatus*, *Quadrula quadrula*) were placed in the casino boat footprint up to the downstream mooring cell (Figure 2-5). All other common and state-listed T&E species were distributed throughout the originally proposed relocation area. Federally endangered species were processed and assigned to grids in the same manner as those collected on the Illinois bank.

### 2.3 Density study

Relocation of unionids from the I-74 bridge footprint provided an opportunity to investigate the effects of increasing resident unionid density at varying rates in relocation areas. Anecdotal evidence suggests that unionid communities often return to pre-relocation densities over time, and that increasing density by a large percentage may be detrimental (Dunn, pers. obs.). To examine the effects of density increases in resident unionid communities, common and state-listed unionids collected at the I-74 bridge were placed in relocation areas in varying numbers, and will be monitored over time.

The proposed density study was set up in 3 different unionid beds (Illiniwek Park, Eagle's Landing, and Upstream; Figure 2-3) to determine if different beds may have different carrying capacities, and to allow results to be replicated. Within each bed, 3 equally-sized areas were established for 3 treatments: a control area in which no unionids were placed, an area in which resident unionid density was to be doubled, and an area in which density was to be tripled (Figures 2-6; 2-7; 2-8). The areas were positioned within each bed based on 3 random points generated with Geospatial Modeling Environment (Beyer, 2012). The size of the areas was determined based on the existing density in each bed (ESI, 2015) and the expected number of unionids to be relocated. The expected number of relocated unionids was calculated as 95% of the total estimated unionids occurring in the Illinois pier areas to ensure that the relocation areas would provide ample capacity if divers collected more unionids than anticipated. Based on these calculations, the size of each study area was initially set at 2,200 m<sup>2</sup>. A circle with a radius of approximately 26 m (85 ft) was generated around each random point to create each treatment area. The circles were moved slightly shoreward or riverward if necessary to ensure the entire study area fell within the previously delineated bed boundary, and in the Eagle's Landing bed, the circles were stretched to fit within the bed boundary while keeping the area constant (Figures 2-6; 2-7; 2-8). Prior to placing relocated unionids in these areas, 80 randomly distributed quantitative samples were collected in each of the 3 treatment areas in the 3 beds (240 samples in each bed; 720 total samples) to estimate density with 15-20% precision.

Samples were collected and unionids were processed as described above.

Data collected in the initial sampling effort resulted in new density estimates that were used to adjust the size of the treatment areas. Density estimates in most of the areas were higher than those observed in 2015 sampling. However, young (<3 years old) and/or small (<25 mm) individuals were extremely abundant in some areas, particularly in the Eagle's Landing bed. Because small individuals take up relatively little space, and few individuals of this size were likely to be collected in the relocation, densities were recalculated to factor out some of these individuals such that the estimated capacity of each area was based more on the age and size classes likely to be relocated. Therefore, densities of unionids  $\geq 3$  years old and of unionids  $\geq 25$  mm in length were calculated for each area. These 2 density estimates were then averaged, and the resulting averaged densities were used to adjust the size of the treatment areas. Based on the new densities, the treatment areas were reduced from 2,200 m<sup>2</sup> to 1,286 m<sup>2</sup>. Additional discussion among ESI biologists suggested that square treatment areas might better facilitate placement of unionids, as the corners of each area could be marked with buoys. The revised treatment areas were thus created as squares within the original circular treatment areas (Figures 2-6; 2-7; 2-8). The number of unionids that could be placed in each area at the prescribed density increase was recalculated using the revised areas, and these estimates were used to guide placement of unionids during the relocation. If the capacity (estimated number of unionids that the area could accommodate) at a particular site was reached during the relocation, use of that site was discontinued.

The double and triple density treatment areas were sampled again immediately following the relocation to quantify changes in density after the addition of relocated unionids. Forty (40) randomly distributed quantitative samples were collected in each treatment area to obtain post-relocation density estimates. Samples were collected and unionids were processed as described above. Density estimates before and after placement of relocated unionids were compared using SYSTAT 13 (Systat Software, San Jose, CA). Data were first tested for normality. Pairwise comparisons were then made using the Wilcoxon signed-rank test. Post-relocation density estimates will be used to assess changes in density over time.

### 3.0 Results and Discussion

#### 3.1 Construction area baseline sampling

##### *Illinois new bridge Action Area*

Baseline sampling of the Illinois new bridge Action Area was conducted on July 7 and 8, 2016. Substrate was very similar to that observed in the 2014 survey. Sand and unionid and zebra mussel shells were the primary substrate constituents throughout most of the area, with some silt and/or gravel present in many samples. Silt and clay were more prevalent at the head of the small island (Figure 3-1). Depth ranged from 0.6 m (2 ft) to 3.7 m (12 ft), and was shallow at the head of the island and deeper in the channels shoreward and riverward.

A total of 327 live unionids of 16 species were collected from the Illinois new bridge Action Area in construction area baseline sampling (Table 3-1). *Quadrula pustulosa* (34.3%), *Obliquaria reflexa* (24.8%), and *Amblema plicata* (15.9%) were by far the most abundant species encountered. With the exception of *Ligumia recta* (5.5%), the remaining species each comprised <3% of the total. An additional 5 species were collected as dead shells. Density throughout the entire Action Area averaged  $13.1 \pm 3.3$  unionids/m<sup>2</sup>. Density within the unionid concentration (from the bank to halfway between Piers 5 and 6) was higher, averaging  $21.8 \pm 4.4$  unionids/m<sup>2</sup>. Nearly half (45.0%) of the individuals collected were  $\leq 5$  years old, and young individuals of 13 species were observed. Observed mortality was 14.6% (Table 3-1). The majority of unionids were collected from the bank to halfway between Piers 5 and 6 (Figure 3-2). Abundance was highest immediately adjacent to the bank and near Pier 5. No unionids were collected riverward of Pier 6 (Figure 3-2).

Observed unionid density was lower in 2016 ( $13.1 \pm 3.3$  unionids/m<sup>2</sup> over the entire Action Area) than in 2014 ( $20.6 \pm 6.6$  unionids/m<sup>2</sup>), but the difference was not significant ( $p > 0.01$ ; Table 3-1). Unionid species composition, community metrics, and distribution varied little among years. *Quadrula pustulosa*, *O. reflexa*, and *A. plicata* were the dominant 3 species in both years, and measures of recruitment and mortality were very similar (Table 3-1). Both sets of data demonstrate that a unionid concentration occurs from the bank to halfway between Piers 5 and 6, and unionid abundance was highest adjacent to the bank in both years. Few or no unionids were collected riverward of Pier 6.

Baseline sampling in the construction area was designed to estimate unionid density within 20-25% of the mean. The study design in the Illinois new bridge Action Area resulted in a density estimate with a 95% confidence interval within 25% of the mean (Table 3-2). Power analysis indicates that 289 and 163 samples would be needed to achieve 15% and 20% precision, respectively.

Unionids were relocated from the direct impact areas at Piers 1-5 in 2016. Additional sampling will be conducted as construction progresses to determine if unionids not relocated from the bridge footprint were impacted by construction. Post-construction sampling will include quantitative sampling to assess changes in community characteristics, and qualitative sampling around newly constructed bridge piers to determine if mortality may have occurred during construction. Monitoring the new bridge footprint with both quantitative and qualitative sampling will continue annually for the first 3 years following construction (2017, 2018, and 2019) and in Years 5, 10, and 15 (2021, 2026, and 2031).



Continued monitoring will seek to describe unionid community changes, particularly mortality, and to document recolonization of unionids in the construction area.

#### *Iowa new bridge Action Area*

Baseline sampling of the Iowa new bridge Action Area was conducted on July 6, 2016. Substrate consisted primarily of gravel, sand, silt, and zebra mussel shells in varying proportions. Some pockets of stable substrate were observed, but in many samples, these constituents were present in a thin layer over bedrock (Figure 3-3). Depth ranged from 2.4 m (8 ft) to 4.9 m (16 ft) and generally increased with distance from the bank.

A total of 161 unionids of 14 species were collected in the Iowa new bridge Action Area in construction area baseline sampling (Table 3-3). *Quadrula pustulosa* (24.2%) and *O. reflexa* (13.7%) were the most abundant species collected; *A. plicata* and *Ellipsaria lineolata* (10.6% each) were also relatively common. The remaining 10 species each comprised <10% of the total. Four (4) additional species were collected as dead shells. Density averaged  $12.9 \pm 3.2$  unionids/m<sup>2</sup>. Recruitment was relatively low; only 12.4% of unionids were  $\leq 5$  years old, and young individuals of only 5 species were observed. Mortality was 12.5% (Table 3-3). Unionid abundance was highest near the shoreward pier, and decreased riverward (Figure 3-4).

Unionid abundance and community characteristics differed somewhat among years. Density was significantly higher in 2016 ( $12.9 \pm 3.2$  unionids/m<sup>2</sup>) than in 2014 ( $3.0 \pm 2.7$  unionids/m<sup>2</sup>;  $p < 0.01$ ), and species richness was also higher in 2016. Observed recruitment was higher in 2014 (22.2%) than in 2016 (12.4%). No mortality was observed in 2014. Unionid distribution was similar in both years; abundance was highest near the bank and decreased riverward. Some of these differences may simply be artifacts of sampling. Unionid distribution was patchy, and samples may have fallen within patches in 2016, and/or outside of patches in 2014. In addition, the sample size used to calculate 2014 metrics was very low, as only 12 samples fell within the new bridge Action Area shoreward of Pier 2 (the same area that was sampled in 2016). Although higher unionid density was observed in 2016, much of the substrate was bedrock, and unionids in this area are likely transient.

Baseline sampling in the construction area was designed to estimate unionid density within 20-25% of the mean. The study design in the Iowa new bridge Action Area resulted in a density estimate with a 95% confidence interval within 25% of the mean (Table 3-2). Power analysis indicates that 134 and 75 samples would be needed to achieve 15% and 20% precision, respectively.

Unionids were relocated from the direct impact areas of all 3 Iowa piers in 2016. As in the Illinois construction area, additional sampling will be conducted as construction progresses to assess impacts of construction on unionids. Sampling and ongoing monitoring will be conducted using the same methods and schedule as the Illinois new bridge Action Area.

### 3.2 Relocation

#### *Illinois bank*

Unionid relocation on the Illinois bank occurred from August 1 to October 25, 2016. Substrate was fairly consistent throughout the search areas, and was primarily composed of gravel, sand, silt, and zebra mussel shells. Numerous dead unionid shells were present on top of and throughout the substrate. Boulder/rip-rap was present on and adjacent to the bank in Pier 1. Silt and clay were more prevalent in the downstream portion of Pier 3 at the head of the small island. Pier 5 fell at the riverward edge of the mussel bed, and substrate in many of the riverward cells was primarily loose sand. Although discharge was unusually high (75,000 – 160,000 cubic feet per second [cfs]), depth generally did not exceed 3.0 m (10 ft).

The relocation effort yielded a total of 139,133 unionids of 32 species (Table 3-4). *Quadrula pustulosa* (35.9%), *O. reflexa* (20.7%) and *A. plicata* (16.0%) were by far the most abundant species encountered, together comprising over 70% of the total. *Ligumia recta* (6.3%) and *Lampsilis cardium* (4.7%) were also relatively common, while the remaining 27 species each comprised <3% of the total. A few species were very rarely encountered; 7 of the 32 total species were each represented by fewer than 10 individuals. Recruitment was observed at all piers and the outfall area. Young individuals of 26 species were collected, and 21.3% of unionids were  $\leq 5$  years old (Table 3-4).

Unionid abundance was highest at the shoreward pier, and declined with distance riverward (Table 3-4). This distributional trend was also observed in previous quantitative sampling of the area in 2014 and 2016. Recruitment followed a similar pattern. However, species richness, species composition, and relative abundance were generally quite similar among all 5 piers (Table 3-4).

Three (3) federally endangered and 4 Illinois threatened species were encountered during the relocation. The federally endangered species *C. monodonta* (n=23), *P. cyphyus* (n=106), and *L. higginsii* (n=747) each comprised a relatively small portion of the total. The Illinois threatened *Cyclonaias tuberculata* (n=1) and *Elliptio dilatata* (n=2) were both quite rare, while *Ellipsaria lineolata* (n=2,640) was moderately common, and *L. recta* (n=8,741) was the fourth most abundant species encountered. *Plethobasus cyphyus*, *L. higginsii*, *E. lineolata*, and *L. recta* were distributed throughout the area and were collected from all 5 piers. *Cumberlandia monodonta* was limited to the first 2 piers and the outfall area, perhaps due to habitat conditions (more boulder/rip-rap present near the bank). *Cyclonaias tuberculata* was only collected in Pier 3, and *E. dilatata* was collected from Piers 3 and 5 (Table 3-4). Neither *C. tuberculata* or *E. dilatata* have been reported live in Pool 15 since 1980, with the exception of 1 *C. tuberculata* recently collected in the Illiniwek Park bed (used as a relocation area; Kelner, 2011; ESI, 2015). *Plethobasus cyphyus*, *L. higginsii*, *E. lineolata*, and *L. recta* were all represented by juveniles as well as adults (Table 3-6); recruitment of *P. cyphyus* has not recently been documented in the Mississippi River. Several species (*Pleurobema sintoxia*, *Lampsilis teres*, *Strophitus undulatus*) listed as threatened or endangered in Iowa but not in Illinois were collected from the Illinois piers as well (Table 3-4).

Common and state T&E species collected at the Illinois piers were distributed among 6 possible relocation areas (Table

3-7). Capacity in the Upstream 2x Density and Illiniwek Park 2x Density sites was reached before the relocation was complete; use of these sites was discontinued after that point. Although unionids were randomly assigned to the relocation areas, the number of unionids placed at each of the remaining 4 sites was similar, ranging from 24,750 to 25,252 (Table 3-7). All *C. monodonta* were placed near previously relocated individuals in Sylvan Slough just downstream of the Rodman Avenue bridge. *Plethobasus cyphus* were randomly distributed among grids at Illiniwek Park and Eagle's Landing, while *L. higginsii* were randomly distributed among grids at Illiniwek Park, Eagle's Landing, and Upstream (Table 3-8). *Cyclonaias tuberculata* and *E. dilatata* were also placed in the *P. cyphus* grids to better track their final placement locations. Three (3) *Lampsilis teres* individuals were placed in the Illiniwek Park *P. cyphus* grid for the same reason, as these individuals were held for a potential genetic sampling project (a fourth *L. teres* was collected late in the relocation and was randomly assigned to one of the relocation areas; Table 3-7). ID numbers of unionids placed in each grid cell are presented in Appendix A. The USFWS Genoa National Fish Hatchery (Genoa) also requested gravid *L. higginsii* and *E. lineolata* for use in their propagation program. Sixty-two (62) gravid *L. higginsii* and 138 gravid *E. lineolata* were provided to Genoa for this purpose (Table 3-8). These individuals were marked in the same manner as other relocated individuals, and will be placed in appropriate relocation areas in conjunction with future I-74 fieldwork once they are done being utilized for propagation. Grids will be sampled annually for the first 2 years after relocation (2017 and 2018), and then at Years 5, 10, and 15 (2021, 2026, and 2031) to assess unionid health and survival.

Original take estimates for the Illinois bank indicated that approximately 282,328 unionids would be relocated from the piers, and an additional 2,931 unionids would be relocated from the outfall area (Stantec, 2016). These estimates were based on the 2014 project area survey, in which samples were spread over a wide area; thus, the sample size within the new bridge Action Area (used to calculate take estimates) was relatively small. The estimated number of unionids to be relocated was therefore re-evaluated using the data collected in the new bridge Action Area in 2016. Based on the new data, approximately 226,193 unionids would be relocated from the pier areas, and 3,143 unionids would be relocated from the outfall area (Table 3-5). The actual number of unionids relocated was lower than expected, as only 137,435 unionids were collected from the pier areas and 1,698 unionids were collected from the outfall area. Although these numbers were outside the 95% confidence interval based on density, sampling to diminishing returns suggests that the relocation goals were achieved in all areas (90% of unionids >1 inch [25 mm] relocated). However, sampling conditions could have contributed to the low numbers. Numerous dead unionid shells were present throughout the substrate, and many live unionids were buried, leading to reduced search efficiency. High current velocity due to unusually high river levels could also have reduced collecting efficiency. A second, less-intensive search of the pier areas will be conducted prior to construction in 2017, which will aid in assessing the success of the relocation.

#### *Iowa bank*

Unionid relocation on the Iowa bank occurred from October 27 to October 31, 2016. The shoreward pier contained the most suitable habitat. Patches of sand, silt, and shell were present among larger areas of bedrock. Substrate in Piers 2 and 3 was primarily bedrock, with some sand and silt often present in a thin layer over the bedrock. Depth throughout the area ranged from approximately 3.0 m (10 ft) to 4.6 m (15 ft).

The relocation effort yielded 1,561 unionids of 20 species (Table 3-9). *Quadrula pustulosa* (17.4%), *O. reflexa* (15.3%), *A. plicata* (15.3%), and *Megaloniaias nervosa* (13.8%) were the most abundant species collected. The remaining 16 species each comprised <10% of the total. Recruitment was low; only 5.8% of unionids were ≤5 years old (Table 3-9). However, young individuals of 14 species were observed.

Unionid abundance was highest in the shoreward pier, where more suitable habitat was present, and declined sharply in the remaining 2 piers. Similar distributional trends were observed in 2014 and 2016 quantitative sampling. Recruitment and species richness also declined with distance riverward. Species relative abundance varied somewhat among piers. *Quadrula pustulosa*, *O. reflexa*, and *A. plicata* were common at all 3 piers. *Megaloniaias nervosa* and *Quadrula quadrula* were both quite abundant in Pier 1, but comprised a smaller percentage of the assemblage in Piers 2 and 3. *Ligumia recta* was moderately common in Piers 1 and 2, but was one of the most abundant species found in Pier 3 (Table 3-9).

Three (3) federally endangered and 1 Iowa threatened species were encountered during the relocation. The federally endangered species, *C. monodonta* (n=1), *P. cyphyus* (n=1), and *L. higginsii* (n=8), were all infrequently encountered, while the Iowa threatened *E. lineolata* (n=100) was moderately common. *Lampsilis higginsii* and *E. lineolata* were both collected at all 3 piers, while *P. cyphyus* was only collected at Pier 1 and *C. monodonta* was only collected at Pier 2 (Table 3-9). No individuals ≤ 5 years old were observed for any of the federally endangered species (although the *P. cyphyus* was only 6 years old and most of the *L. higginsii* were ≤10 years old), but several juvenile *E. lineolata* were collected (Table 3-11).

Common and state T&E species collected in the Iowa piers were relocated to the area along the Iowa bank just upstream of the new bridge corridor. Pre-relocation quantitative sampling of the relocation area was conducted on July 6 and 7, 2016. Substrate consisted of sand, silt, clay, and shell in varying proportions. Shell was more common in the downstream portion of the area, while the upstream portion was dominated by silt and clay. Patches of bedrock were observed in several samples, primarily in the riverward portion of the area (Figure 3-5). Depth ranged from 0.6 m (2 ft) to 4.6 m (15 ft). The upstream shoreward corner was shallowest, and depth increased riverward and downstream.

Density in the Iowa relocation area averaged  $4.6 \pm 1.4$  unionids/m<sup>2</sup> (Table 3-12). Approximately 37,034 unionids could be placed in this area at a 50% density increase, indicating the area was large enough to accommodate the expected number of unionids relocated from the Iowa piers. Unionid abundance was highest in the downstream portion of the area, and decreased upstream where habitat contained more silt and clay (Figure 3-6). Although community composition differed slightly, most of the species collected in the Iowa construction area were also collected in the proposed relocation area, suggesting that this area would serve as a suitable recipient site for unionids relocated from the Iowa piers (Table 3-12). Several of the most common and/or tolerant species (*A. plicata*, *L. cardium*, *L. fragilis*, *P. alatus*, *Q. quadrula*) were placed in the old casino boat footprint, while all other species were distributed throughout the originally defined relocation area (see Figure 2-5). Federally endangered species were placed in the same sites/grids as those collected on the Illinois bank (Table 3-8).

Original take estimates for the Iowa bank indicated that approximately 10,663 unionids would be relocated from the Iowa piers (Stantec, 2016). As on the Illinois bank, these estimates were based on the 2014 project area survey, and the sample size within the new bridge Action Area (used to calculate take estimates) was quite small. The estimated number of unionids to be relocated was re-evaluated with data collected in the new bridge Action Area in 2016. However, samples in 2016 were only collected shoreward of Pier 2. Therefore, estimated number of relocated unionids was only calculated for Piers 1 and 2, where the 2016 data would be relevant. Based on the new data, approximately 18,045 unionids would be relocated from Pier 1 and 17,689 unionids would be relocated from Pier 2 (Table 3-10). The actual number of relocated unionids was much lower than expected in both areas, as only 1,014 and 411 unionids were collected from Pier 1 and Pier 2, respectively. This discrepancy may be due in part to the variability in unionid habitat and distribution on the Iowa bank. Samples collected in 2016 may have fallen in more favorable patches of habitat as compared to 2014 samples, potentially overestimating density in the area. In addition, the Iowa piers were not gridded off and searched to diminishing returns, and high flow and large patches of bedrock may have caused divers some difficulty in maintaining position, reducing their search efficiency. Both of these factors may have resulted in some unionids being missed. Unionid abundance at Pier 3 was extremely low in 2014, and substrate was primarily bedrock. Few unionids were expected in this pier, and indeed, only 136 individuals were collected in the relocation effort. A second, less-intensive search of the pier areas will be conducted prior to construction in 2017.

### 3.3 Density study

#### *Illiniwek Park*

Pre-relocation sampling at Illiniwek Park was conducted July 6 to 8, 2016. The Control and 3x Density areas were located in close proximity to each other, and featured similar habitat. Substrate was primarily a mixture of silt and clay, and depth rarely exceeded 1.8 m (6 ft; Table 3-13). The 2x Density area was located farther downstream, where habitat was somewhat different. Substrate primarily consisted of sand, silt, clay, and shell in varying proportions, and depth ranged from 1.8 m (6 ft) to 3.7 m (12 ft; Table 3-13).

Unionid density and species composition varied among the 3 treatment areas. Density was lowest in the control area ( $10.8 \pm 1.9$  unionids/m<sup>2</sup>), intermediate in the 2x Density area ( $20.0 \pm 2.5$  unionids/m<sup>2</sup>), and highest in the 3x Density area ( $28.8 \pm 4.9$  unionids/m<sup>2</sup>; Table 3-14). Species composition in the control and 3x Density areas was similar. *Amblema plicata* and *O. reflexa* dominated the catch in both areas, with most other species representing <5% of the total. *Quadrula pustulosa* was the most abundant species in the 2x Density area, though *A. plicata* and *O. reflexa* were also common. Species richness was lowest in the control area and highest in the 2x Density area. Recruitment was slightly higher in the control and 3x Density areas (57.7% and 53.0%, respectively) than in the 2x Density area (45.4%). Mortality was low in all 3 areas, ranging from 0.7% to 2.3% (Table 3-14).

Densities from pre-relocation sampling were used to determine how large the 2x and 3x Density areas needed to be to achieve the desired density increases. Densities were adjusted to factor out some of the youngest and smallest individuals; based on adjusted densities, the 2x Density area could accommodate 23,791 relocated unionids, and the 3x

Density area could accommodate 57,613 unionids (Table 3-15). This target was reached in the 2x Density area; 23,694 unionids were placed. However, only 24,837 unionids were placed in the 3x Density area, falling short of the estimated number of unionids needed to triple the existing density.

The 2x and 3x Density areas were sampled again on November 4, 2016. Based on the total number of unionids placed, density in the 2x Density area was expected to increase 99.6%, and density in the 3x Density area was expected to increase 86.2% (Table 3-15). The observed percent increase in density in both areas was lower than expected; density was increased 74.0% in the 2x Density area and 85.8% in the 3x Density area. In addition, nearly half of the individuals collected were marked (relocated) individuals, suggesting that placement of relocated unionids nearly doubled the resident density in both areas (Table 3-14). However, overall density estimates were lower than expected. Density of resident (unmarked) unionids decreased significantly between sampling events, averaging only  $12.7 \pm 3.4$  unionids/m<sup>2</sup> in the 2x Density area and  $12.0 \pm 3.3$  unionids/m<sup>2</sup> in the 3x Density area after the relocation (Table 3-14; Table 3-15). Based on the number of unionids placed and the size of each area, density of relocated unionids should have increased to 18.4 unionids/m<sup>2</sup> in the 2x Density area and 19.3 unionids/m<sup>2</sup> in the 3x Density area (Table 3-15). However, observed densities of relocated unionids were only  $9.4 \pm 4.2$  unionids/m<sup>2</sup> and  $10.3 \pm 5.3$  unionids/m<sup>2</sup> in the 2x and 3x Density areas, respectively. The decrease in resident density, coupled with the low density of relocated unionids, suggests that both resident and relocated unionids may have emigrated horizontally out of the sampled area between sampling events, or burrowed such that they were not collected during sampling. It is not known whether movement to this degree is common, as most unionid monitoring studies involve sampling only once per year, rather than twice (as was the case in this study). Variation in the depth to which samples were excavated is also a factor that may have influenced the results, as different divers were used in each sampling event; quadrats excavated to shallower depths could result in missing some unionids. However, Illiniwek Park was sampled last in the second sampling event, by which time divers had been instructed to excavate more substrate in samples. Significant decreases in density were still observed at this site even after divers began to excavate more substrate, suggesting that sample depth was not the primary factor explaining the density decrease.

Species composition was generally similar in both areas before and after the relocation. Species richness of resident unionids was lower after the relocation, but this may be due in part to the smaller sample size (40 samples post-relocation vs. 80 samples pre-relocation). Total species richness, including relocated unionids, was similar to pre-relocation richness in both the 2x and 3x Density areas. Recruitment of resident unionids was similar before and after the relocation; 52.0% of unionids in the 2x Density area and 59.8% of unionids in the 3x Density area were  $\leq 5$  years old. Juveniles comprised a smaller percentage of relocated unionids (18.1% and 16.0% in the 2x and 3x Density areas, respectively), but this was expected, given that the majority of unionids relocated from the bridge were  $>5$  years old. Mortality of resident unionids increased to 19.1% in the 2x Density area and 9.3% in the 3x Density area. Some mortality (2.9%) of relocated unionids was observed in the 3x Density area (Table 3-14); this estimate is well within the estimated 10% mortality due to relocation. No mortality of relocated unionids was observed in the 2x Density area. One (1) *L. higginsii* marked with a slash mark but no ID number was encountered in the 2x Density area. This individual was

subsequently given a unique ID number and moved to the Illiniwek Park *L. higginsii* grid (see Table 3-8).

### *Eagle's Landing*

Pre-relocation sampling at Eagle's Landing was conducted July 8 to 12, 2016. As at Illiniwek Park, the Control and 3x Density areas were located in close proximity to each other, and were characterized by similar habitat. Substrate was primarily composed of silt and zebra mussel shells, transitioning to sand on the far riverward edge of the area (Table 3-13). Cobble was the dominant substrate constituent in samples immediately adjacent to the bank. Depth in both areas ranged from 0.3 m (1 ft) near the bank to 2.7 m (9 ft) riverward. The 2x Density area was located farther downstream, but habitat was generally similar to the other areas. Substrate throughout most of the area was a mixture of sand, silt, and shell, with more silt and clay present on the shoreward edge, and sand on the riverward edge. Depth ranged from 0.3 m (1 ft) near shore to 3.7 m (12 ft) riverward (Table 3-13).

Pre-relocation density varied, but was high in all study areas. Density was lowest in the 2x Density area ( $61.5 \pm 10.2$  unionids/m<sup>2</sup>), intermediate in the control area ( $78.0 \pm 15.9$  unionids/m<sup>2</sup>), and highest in the 3x Density area ( $91.8 \pm 19.8$  unionids/m<sup>2</sup>; Table 3-16). Species composition was generally similar among study areas. *Utterbackia imbecillis*, *A. plicata*, *Q. pustulosa*, and *O. reflexa* were the most abundant species in all areas, though their relative abundance differed slightly. *Leptodea fragilis* was also common in the control and 3x Density areas. Species richness was lowest in the 2x Density area (20 species) and highest in the 3x Density area (24 species). The majority (75.5% - 78.6%) of unionids collected in the study areas were  $\leq 5$  years old. Mortality was low, ranging from 4.0% (3x Density area) to 5.8% (2x Density area; Table 3-16).

Adjusted densities (factoring out young and small individuals) were used to calculate capacity in the 2x and 3x Density areas. Because *U. imbecillis* was so common, and most individuals of this species were young and/or small, densities were significantly reduced. Based on adjusted densities, 61,471 unionids could be placed in the 2x Density area, and 177,725 additional unionids could be placed in the 3x Density area (Table 3-15). The number of unionids collected in the relocation effort was less than anticipated, so these targets were not met. A total of 25,124 unionids were placed in the 2x Density area, and 25,252 unionids were placed in the 3x Density area (Table 3-15).

The 2x and 3x Density areas were sampled again on November 3, 2016. Density increases of 40.9% and 28.4% were expected for the 2x Density and 3x Density areas, respectively. The observed percent increase was lower than expected in both the 2x Density (31.4%) and 3x Density (17.2%) areas (Table 3-15). However, overall density estimates were much lower than expected. Resident unionid density declined significantly to  $24.2 \pm 5.0$  unionids/m<sup>2</sup> in the 2x Density area and  $36.1 \pm 8.7$  unionids/m<sup>2</sup> in the 3x Density area after the relocation ( $p < 0.01$ ; Table 3-15; Table 3-16). Based on the number of unionids placed, density of relocated unionids should have increased to 19.5 unionids/m<sup>2</sup> in the 2x Density area and 19.6 unionids/m<sup>2</sup> in the 3x Density area (Table 3-15). However, observed density of relocated unionids was only  $7.6 \pm 3.6$  unionids/m<sup>2</sup> in the 2x Density area and  $6.2 \pm 3.9$  unionids/m<sup>2</sup> in the 3x Density area (Table 3-15; Table 3-16). As at Illiniwek Park, this information suggests that movement (either horizontal or vertical) of both resident and

relocated unionids occurred between sampling events. Variation in the depth to which samples were excavated could have contributed to the density changes as well, although this does not appear to have been the primary issue. Seasonal fluctuation in the abundance of young individuals was also considered. *Utterbackia imbecillis*, represented almost exclusively by young and/or small individuals, was very commonly encountered in pre-relocation sampling, but was rare in the second sampling event. It was initially thought that the apparent decline in this species may have contributed to the decrease in density. However, the overall percentage of individuals  $\leq 5$  years old did not change appreciably between sampling events, suggesting that a change in the abundance of juveniles was not a factor.

Species composition differed somewhat among sampling events. *Utterbackia imbecillis* was one of the most commonly encountered species in pre-relocation sampling, but was scarce in post-relocation samples. However, relative abundance of most other species was similar. Species richness was lower in post-relocation samples, but this may be due to smaller sample size. Recruitment of resident unionids was similar in pre- and post-relocation samples; juveniles comprised 66.1% and 61.8% of unionids in the 2x and 3x Density areas, respectively. As expected, recruitment was much lower among relocated unionids than resident unionids (Table 3-16). Mortality of resident unionids in both the 2x Density (14.2%) and 3x Density (8.4%) areas increased somewhat over pre-relocation values. Mortality of relocated unionids was 5.0% in the 2x Density area and 3.1% in the 3x Density area, well within the estimated 10% mortality due to relocation (Table 3-16).

### *Upstream*

Pre-relocation sampling at Upstream was conducted July 8 to 9 and July 12 to 13, 2016. Habitat was generally similar among the 3 study areas. Substrate was primarily composed of sand, silt, and zebra mussel shells, with smaller amounts of cobble, gravel, and clay present in some samples (Table 3-13). The shoreward edge of the 3x Density area fell on a depositional area adjacent to the bank, where substrate was a mixture of silt and clay. Depth increased with distance from the bank, reaching a maximum of 13 ft (4.0 m; Table 3-13).

Pre-relocation density was similar in the 2x and 3x Density areas ( $11.8 \pm 2.2$  unionids/m<sup>2</sup> and  $11.1 \pm 2.4$  unionids/m<sup>2</sup>, respectively), and higher in the control area ( $17.8 \pm 2.6$  unionids/m<sup>2</sup>; Table 3-17). Species composition and richness were similar among all areas. *Amblema plicata*, *Q. pustulosa*, and *O. reflexa* were the most commonly encountered species, though their relative abundance differed in each area. Fifteen (15) species were collected in the 2x and 3x Density areas, and 16 species were collected in the control area. Recruitment ranged from 40.0% (2x Density area) to 59.3% (3x Density area). Observed mortality was moderate in the control and 2x Density areas (13.2% and 17.0%, respectively) and highest in the 3x Density area (31.8%; Table 3-17).

Adjusted densities (factoring out young and small individuals) were used to calculate capacity in the 2x and 3x Density areas. Based on adjusted densities, 14,017 unionids could be placed in the 2x Density area, and 25,720 unionids could be placed in the 3x Density area (Table 3-15). This target was met for the 2x Density area; 14,457 unionids were placed. The 3x Density area fell slightly short of the target, as only 24,750 unionids were placed (Table 3-15).



The 2x and 3x Density areas were sampled again on November 1, 2016. Density was expected to increase 103.1% and 192.5% in the 2x Density and 3x Density areas, respectively. Actual percent changes in density were lower than expected; increases of only 53.4% and 75.0% were observed in the 2x and 3x Density areas, respectively (Table 3-15). Similarly, overall density of relocated unionids was lower than anticipated. Based on the number of unionids placed, density of relocated unionids should have increased to 11.2 unionids/m<sup>2</sup> in the 2x Density area and 19.2 unionids/m<sup>2</sup> in the 3x Density area (Table 3-15). However, observed density of relocated unionids was only  $4.7 \pm 1.9$  unionids/m<sup>2</sup> in the 2x Density area and  $8.1 \pm 3.2$  unionids/m<sup>2</sup> in the 3x Density area (Table 3-15; Table 3-17). In contrast to the other 2 beds, however, resident unionid density did not change significantly, averaging  $8.8 \pm 2.4$  unionids/m<sup>2</sup> in the 2x Density area and  $10.8 \pm 3.4$  unionids/m<sup>2</sup> in the 3x Density area after the relocation (Table 3-15; Table 3-17). This suggests that some relocated unionids appear to have emigrated from the sampled areas, but resident unionids remained.

Species composition and richness were similar before and after the relocation, though species richness was slightly lower for relocated unionids than for residents (Table 3-17). Recruitment of resident unionids increased to 73.9% in the 2x Density area, but was similar to pre-relocation values (46.3%) in the 3x Density area. As in the other beds, juveniles comprised a smaller percentage of relocated unionids; 23.4% of relocated unionids in the 2x Density area and 8.6% in the 3x Density area were  $\leq 5$  years old. Mortality of resident unionids was similar among sampling events, ranging from 24.1% in the 2x Density area to 35.7% in the 3x Density area after the relocation. Mortality of relocated unionids was observed in both areas, but was well within the estimated 10% mortality due to relocation (Table 3-17).

Post-relocation sampling of the density study areas yielded variable results. Several areas (Illiniwek Park 2x Density, Upstream 2x Density, Upstream 3x Density) met or nearly met the target number of individuals needed to double or triple initial density. However, observed percent increases in density were generally lower than expected (Table 3-15). Distribution of relocated unionids throughout the relocation areas was not uniform. Although efforts were made to spread unionids throughout the area, relocated unionids appeared to be concentrated in the center of the placement areas, as more marked individuals were recovered near the center than near the boundaries. Thus, sampling may have underestimated the density of relocated unionids.

In addition to the low percent increase in relocated unionids, post-relocation density of resident unionids declined significantly in the Illiniwek Park and Eagle's Landing treatment areas. The decline does not appear to be due to a loss of young individuals, as recruitment in resident unionids was generally similar before and after the relocation. Rather, it appears that both resident and relocated unionids emigrated horizontally from the sampled areas, or burrowed such that they were not detected during sampling. Pre-relocation sampling was conducted in July, while post-relocation sampling occurred in November. Water temperature had decreased significantly by post-relocation sampling, and unionids may have been buried deeper, resulting in fewer unionids collected if samples were dug to the same depth as those in pre-relocation sampling. Mississippi River discharge was unusually high throughout the summer and fall of 2016, and high discharge may have dislodged some unionids, both resident and relocated, from the substrate. Relocated unionids were generally released in the upstream half of the areas to offset increased drift due to high discharge. Although this effort

was made to ensure relocated unionids settled to the bottom within the study areas, some unionids may have drifted downstream when they were released and/or before they were able to burrow into the substrate. Variation in sample depth could also have contributed to the observed changes in density; however, the data suggest that this was not the primary factor. The Upstream site was sampled first, followed by Eagle's Landing and Illiniwek Park. Divers were instructed to excavate additional substrate after sampling at Eagle's Landing was completed. No significant changes in resident density were observed at Upstream, before divers began excavating deeper, but significant decreases in density were observed at Illiniwek Park even with increased excavation, suggesting that sample depth was not the primary factor influencing the observed density changes.

Monitoring these areas in future years will provide additional data to explain the density changes observed in 2016. All density study areas will be sampled again in the 2017 field season. Results that are more similar to pre-relocation densities would suggest some sort of seasonal movement (burrowing) or other temporary fluctuation. However, results that are similar to post-relocation densities would suggest that unionids moved out of the study area, either actively or passively.

Although approximately 138,000 unionids were placed in the 6 density study areas, post-relocation sampling suggests that density was not increased to desired levels in any of the areas. Study areas will be monitored in the future to quantify changes in density over time. Random quantitative samples will be collected as in the initial sampling event. Sampling will occur annually for 2 years following the relocation (2017 and 2018), and at Years 5, 10, and 15 (2021, 2026, 2031). Data collected in monitoring events will be compared to 2016 pre- and post-relocation data to better assess whether density was actually changed by placement of relocated individuals, and to observe trends in density over time.

#### 4.0 Summary and Conclusions

Unionid sampling for the I-74 bridge project in 2016 included baseline sampling of bridge construction areas, relocation of unionids from direct impact areas in the new bridge corridor, and pre- and post-relocation sampling of relocation areas. Results of sampling in the construction areas were generally similar to results from the initial project area survey (ESI, 2014), though density on the Iowa bank was higher than previously observed. These samples provided information on unionid density and community characteristics that can be compared to data collected in future years to assess the effects of construction on unionid communities in the new bridge corridor.

A total of 140,694 unionids of 32 species were collected, marked, and relocated from direct impact areas in the new bridge corridor. A few common species (*A. plicata*, *Q. pustulosa*, *O. reflexa*) comprised the majority of relocated unionids, but 3 federally endangered species (*C. monodonta*, *P. cyphus*, and *L. higginsii*) and several additional state T&E species were collected as well. Federally endangered species were placed in grids, which will be monitored periodically to assess health and survival of relocated individuals. Common and state T&E species collected on the Illinois bank were distributed among 6 potential relocation areas, while those collected on the Iowa bank were placed in a single relocation area just upstream of the new bridge corridor.

Common and state T&E species collected on the Illinois bank were used in a study designed to assess the effects of increasing unionid density by varying degrees. Control, 2x Density, and 3x Density areas were established in each of 3 previously delineated unionid beds, and the treatment areas were sampled prior to the relocation to assess initial unionid density and community characteristics. Unionids collected and relocated from the Illinois piers were placed in the 2x and 3x Density areas, and these areas were sampled again after the relocation was complete. Although numerous unionids were placed in these areas, densities did not increase as expected. Several factors, including distribution of relocated unionids, temporal trends in community composition, and high river levels, may explain some of the results. Future monitoring of these areas will allow for better assessment of density trends over time.

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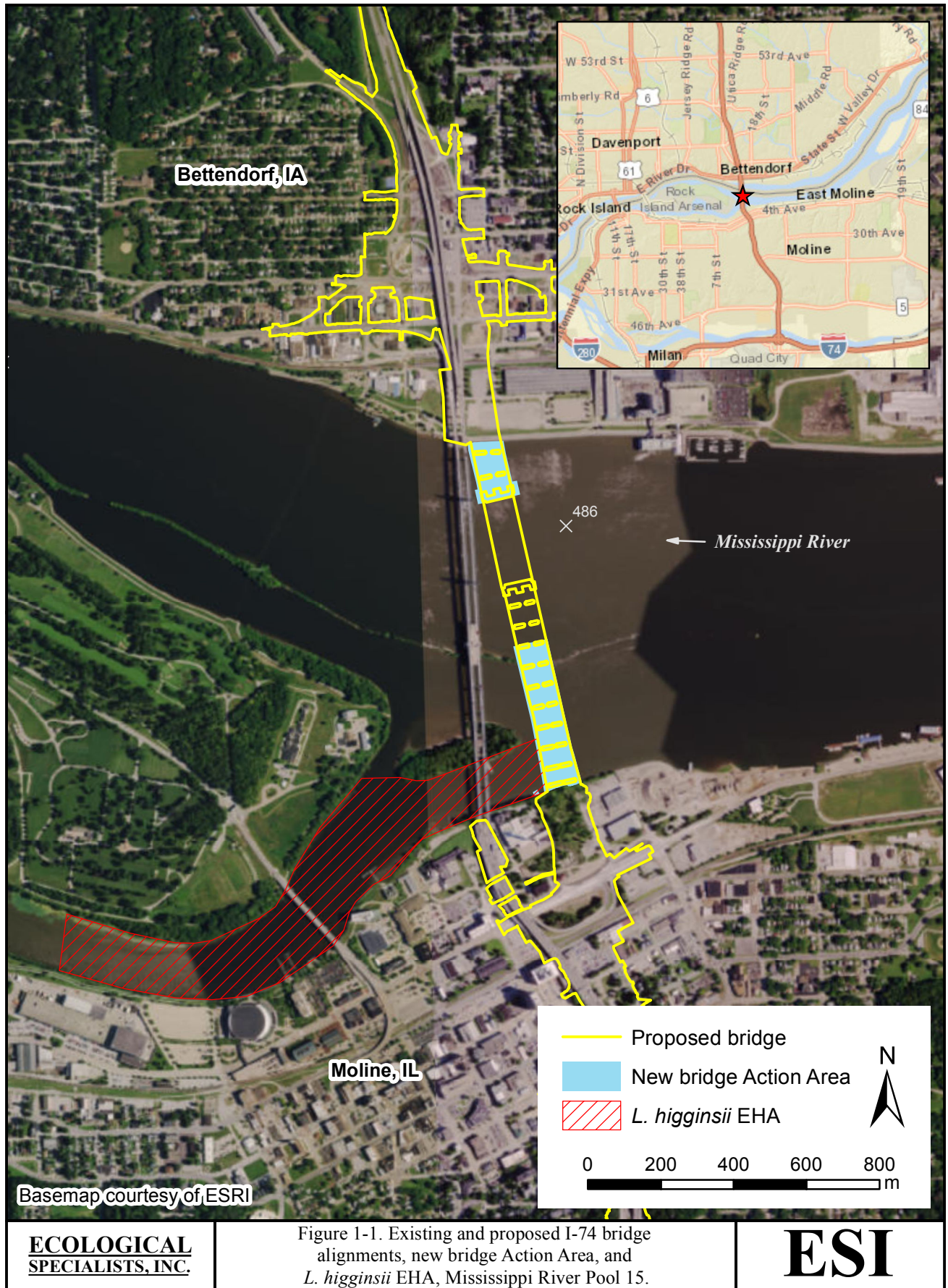
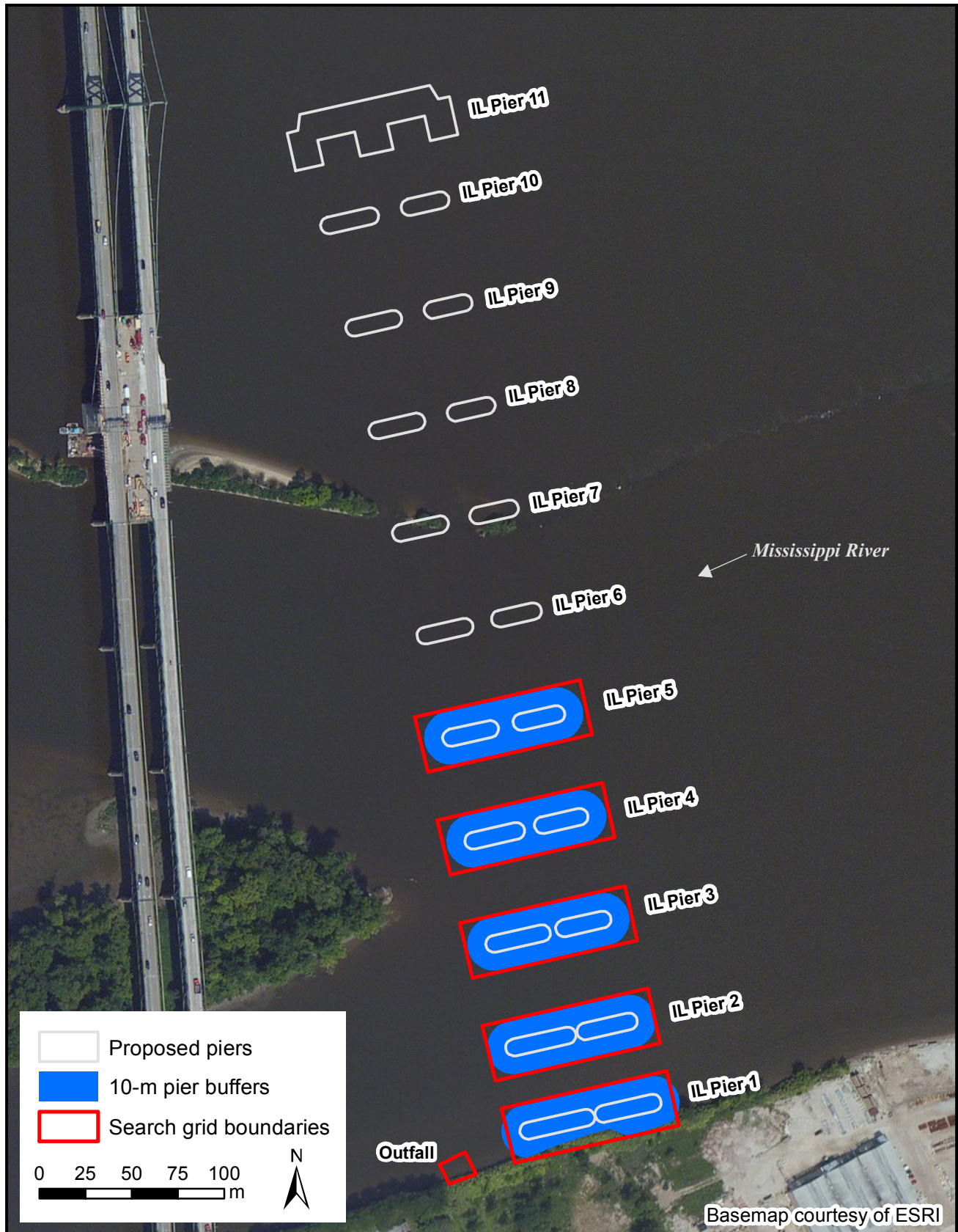


Figure 1-1. Existing and proposed I-74 bridge alignments, new bridge Action Area, and *L. higginsii* EHA, Mississippi River Pool 15.

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Figure 2-1. Proposed pier locations and relocation search grid boundaries on the Illinois bank, August - October 2016.

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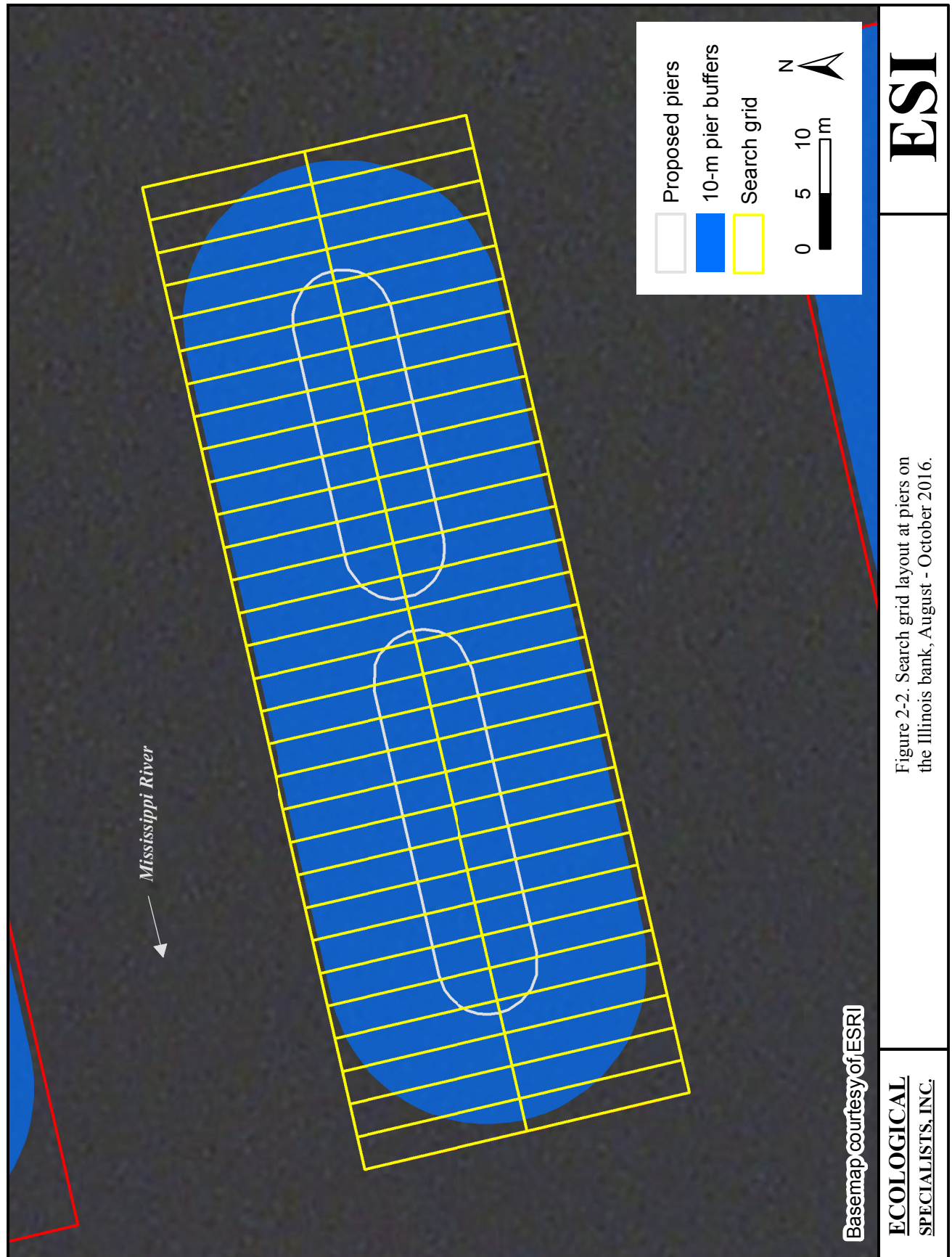


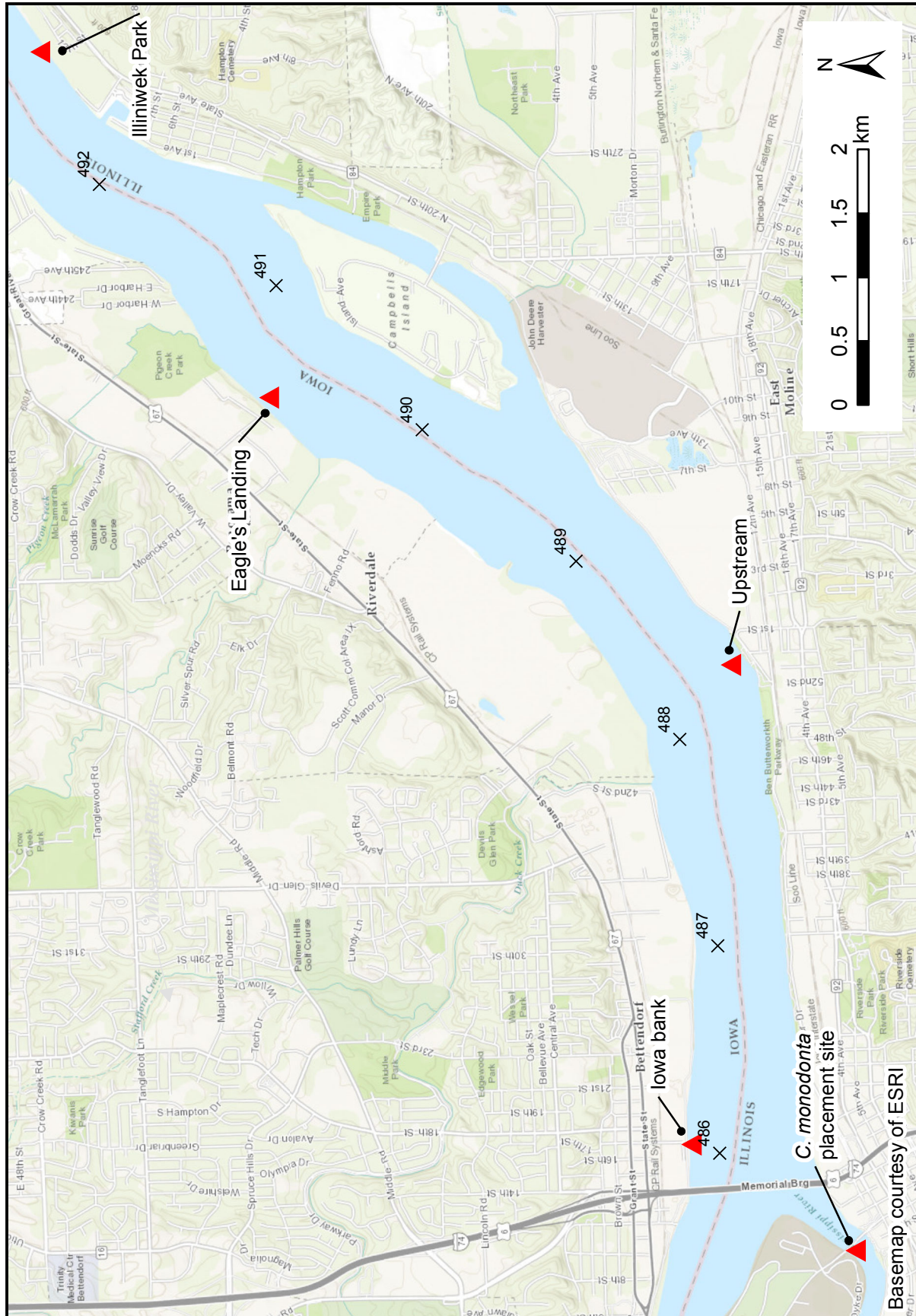
Figure 2-2. Search grid layout at piers on the Illinois bank, August - October 2016.

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Basemap courtesy of ESRJ

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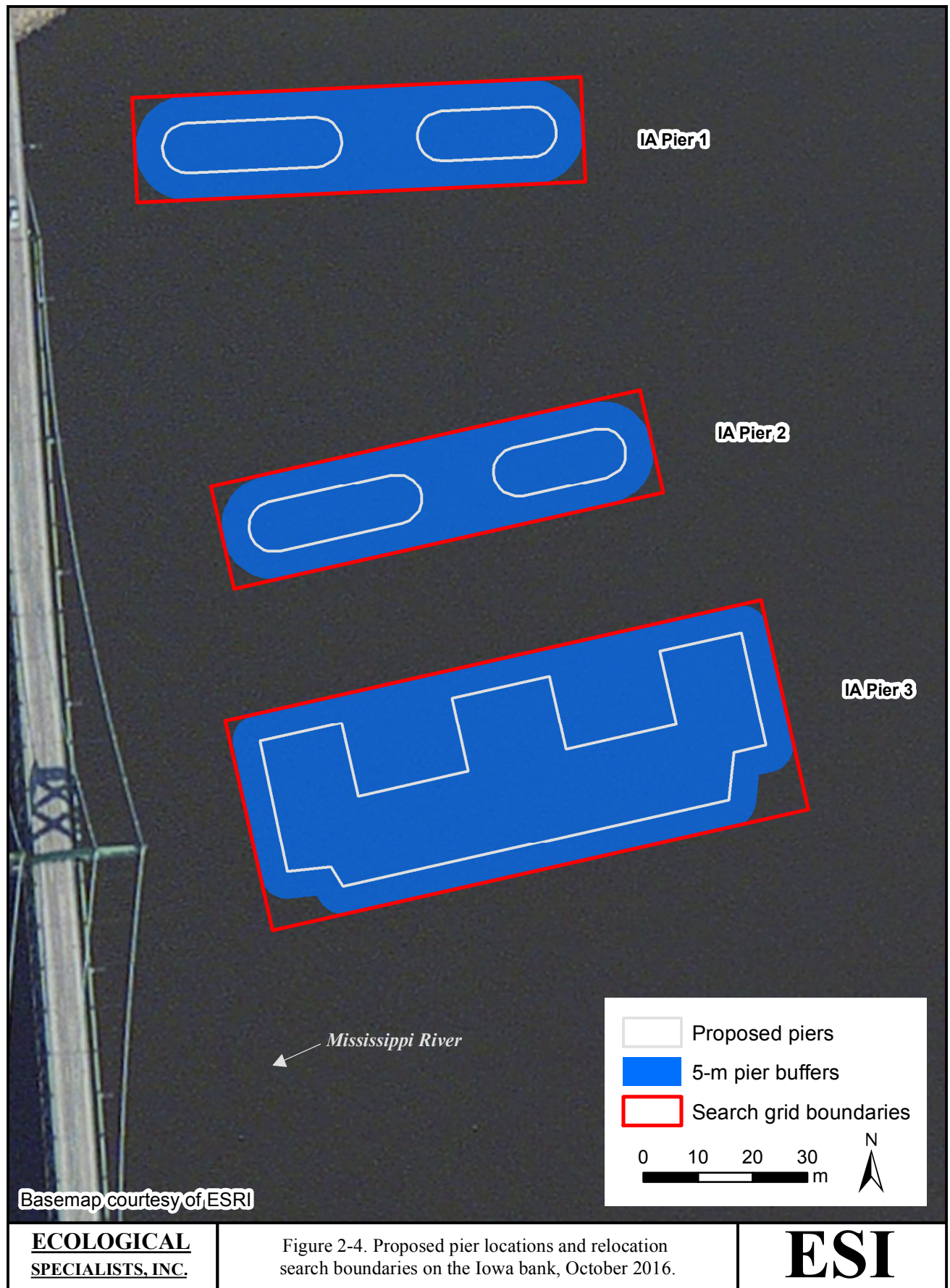
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Figure 2-3. Relocation areas for unionids collected from the new bridge footprint, August - October 2016.

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Basemap courtesy of ESRI







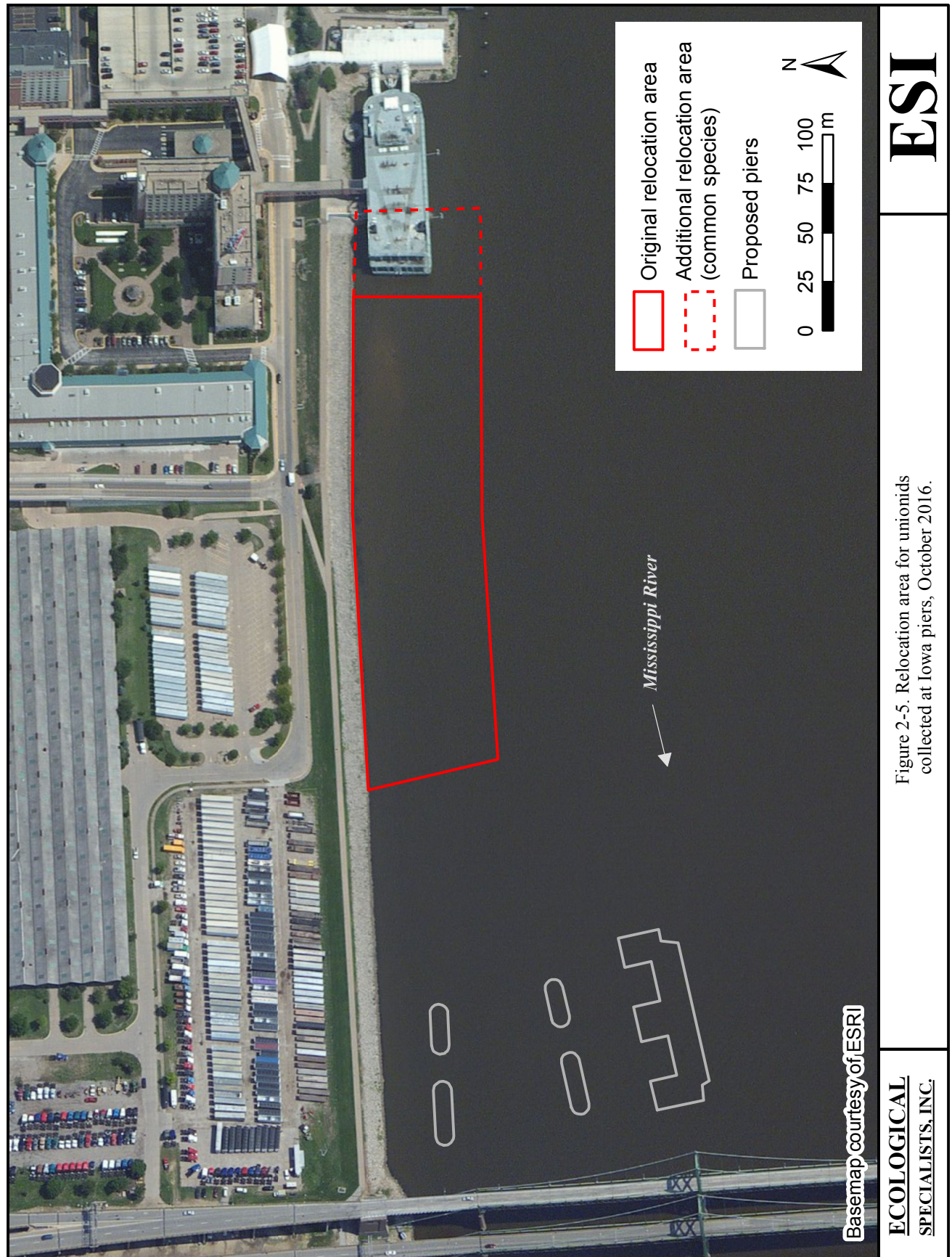


Figure 2-5. Relocation area for unionids collected at Iowa piers, October 2016.



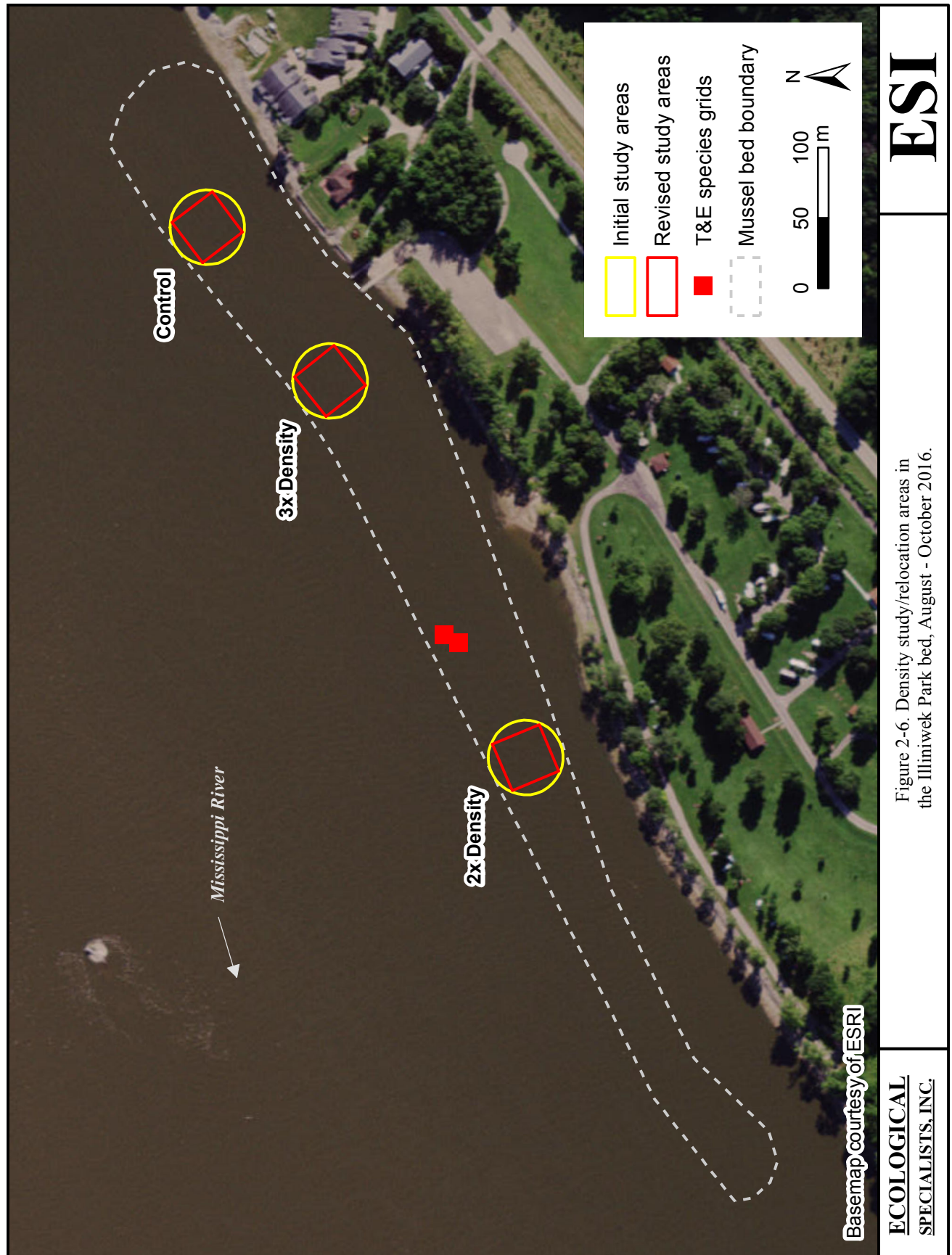
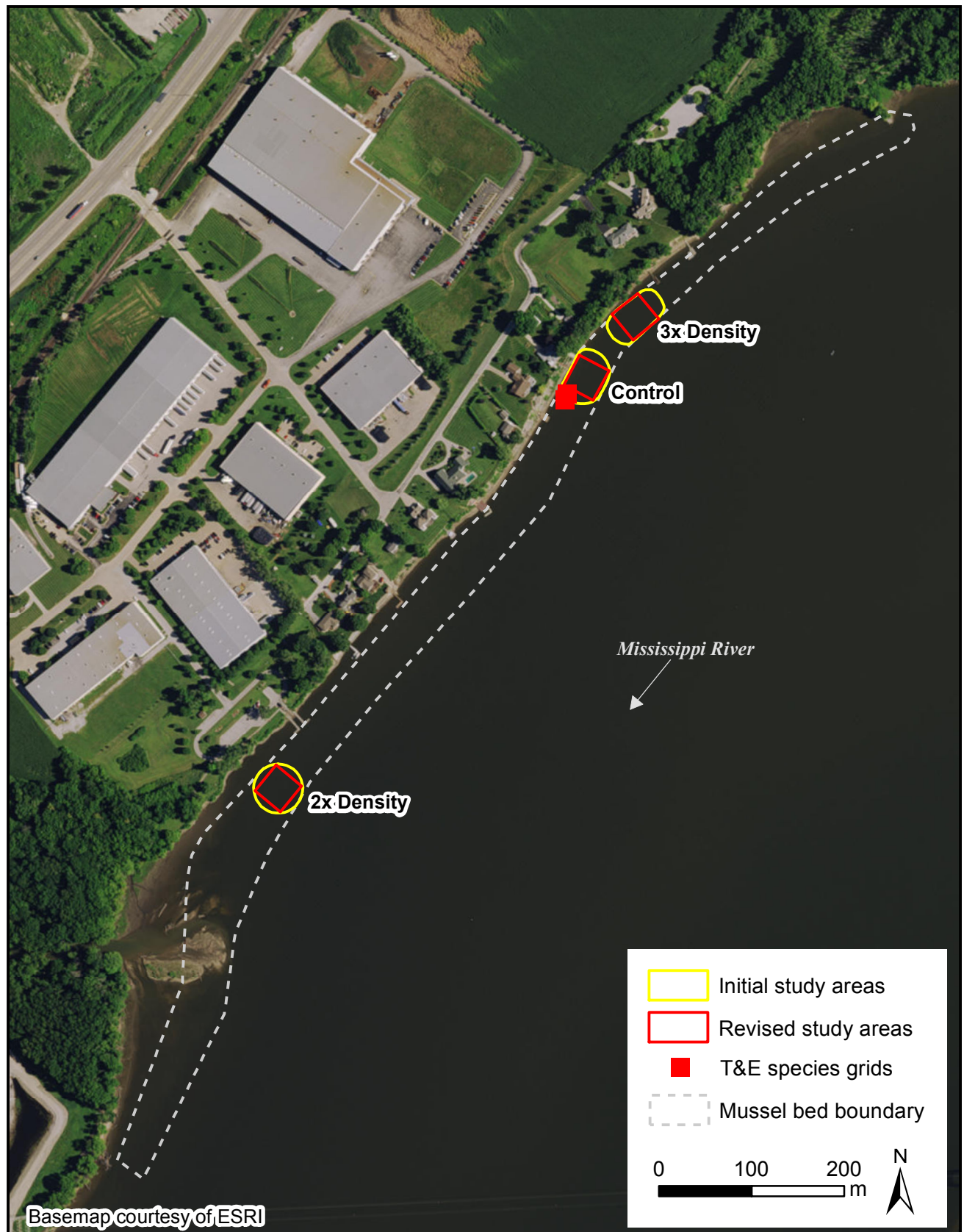


Figure 2-6. Density study/relocation areas in the Illiniwek Park bed, August - October 2016.



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Figure 2-7. Density study/relocation areas in the Eagle's Landing bed, August - October 2016.

**ESI**



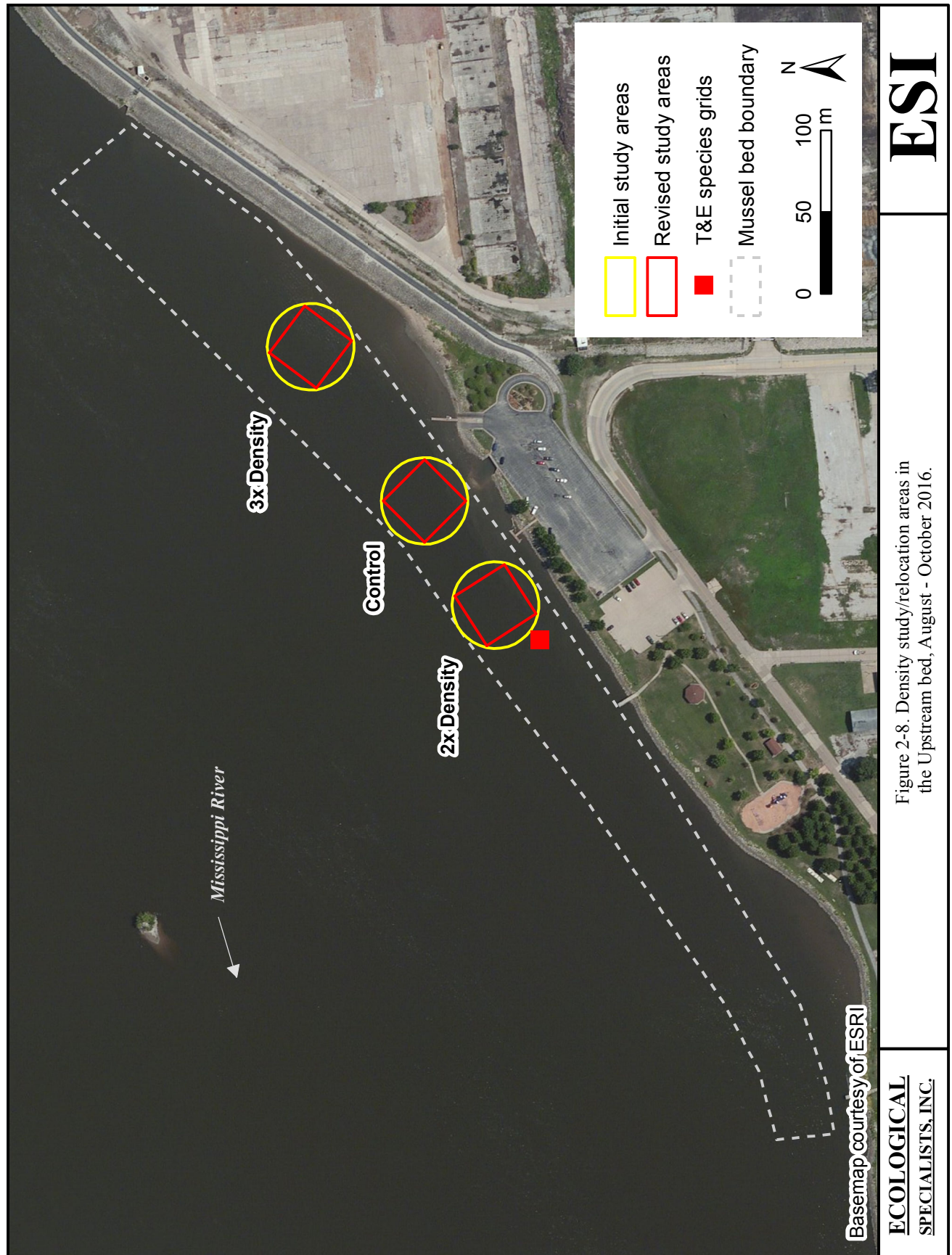
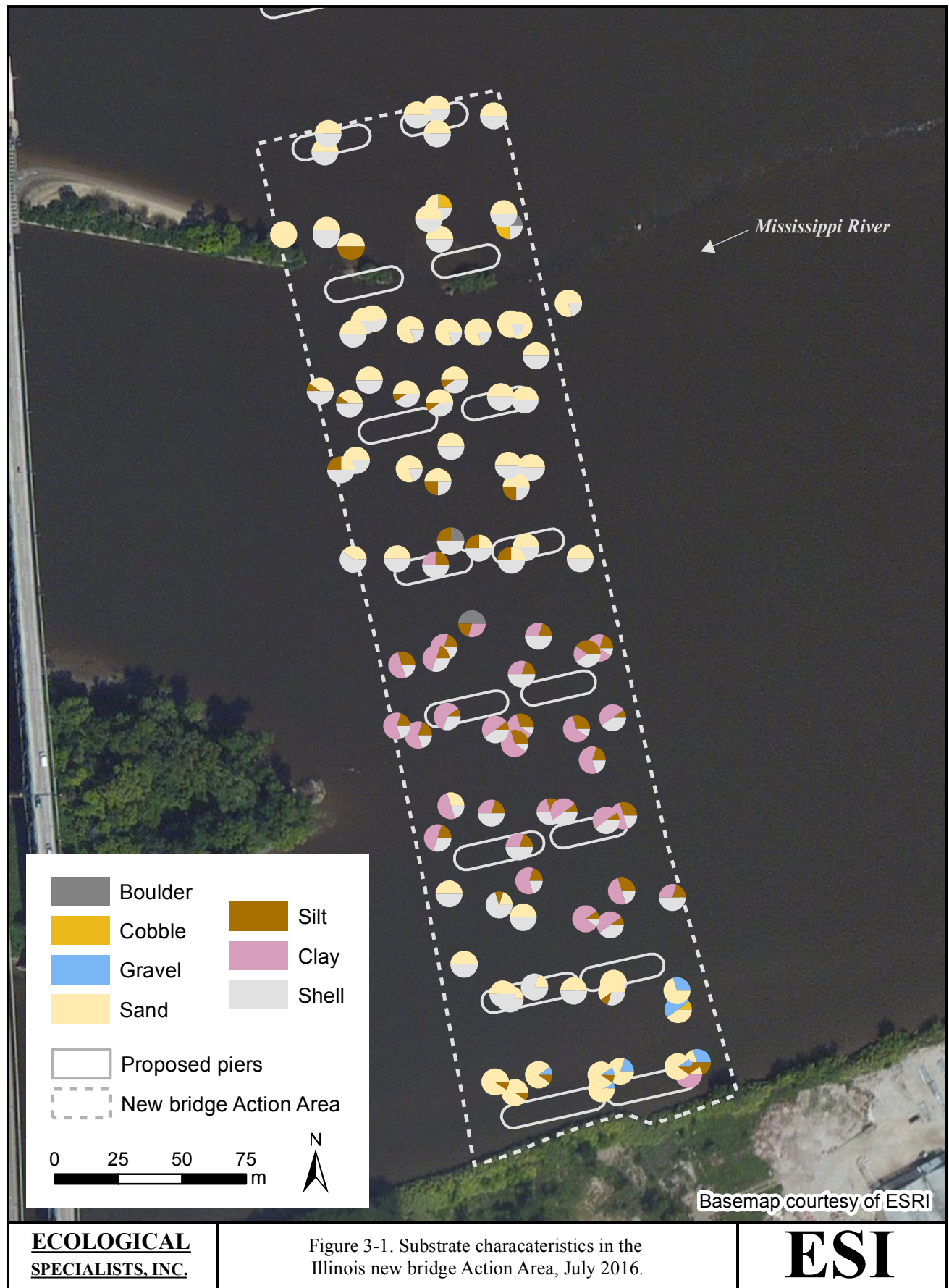
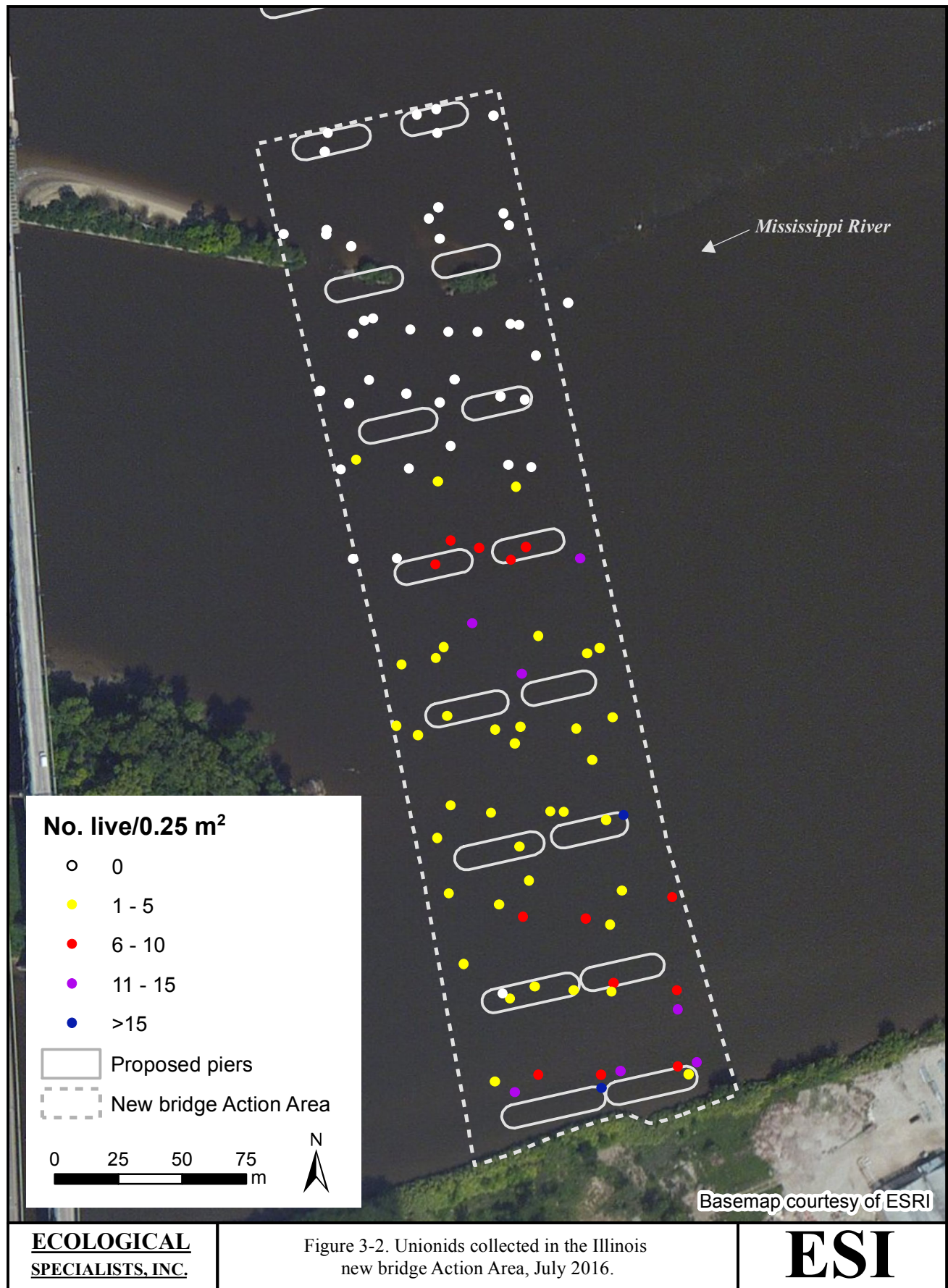


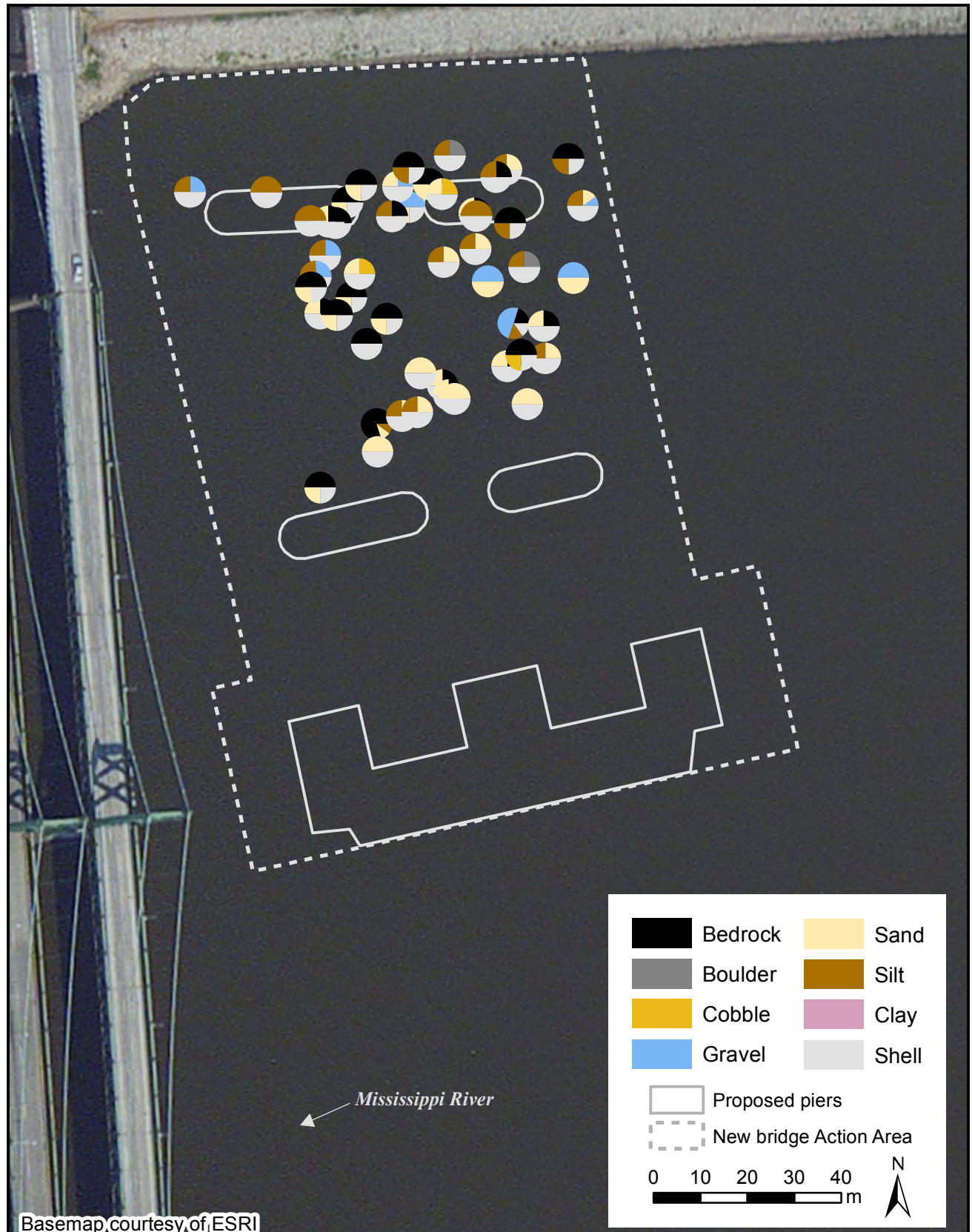
Figure 2-8. Density study/relocation areas in the Upstream bed, August - October 2016.











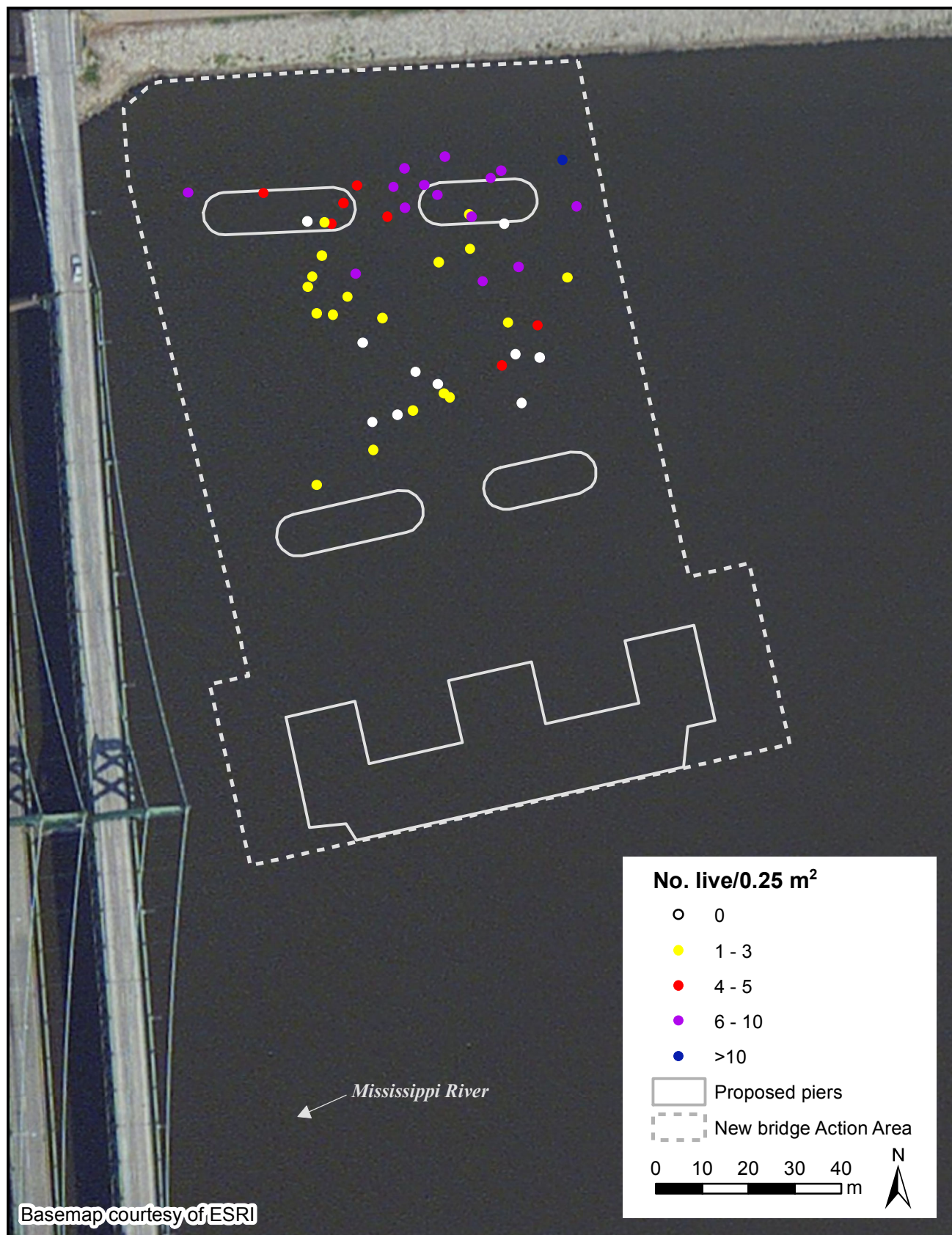
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Figure 3-3. Substrate characteristics in the Iowa new bridge Action Area, July 2016.

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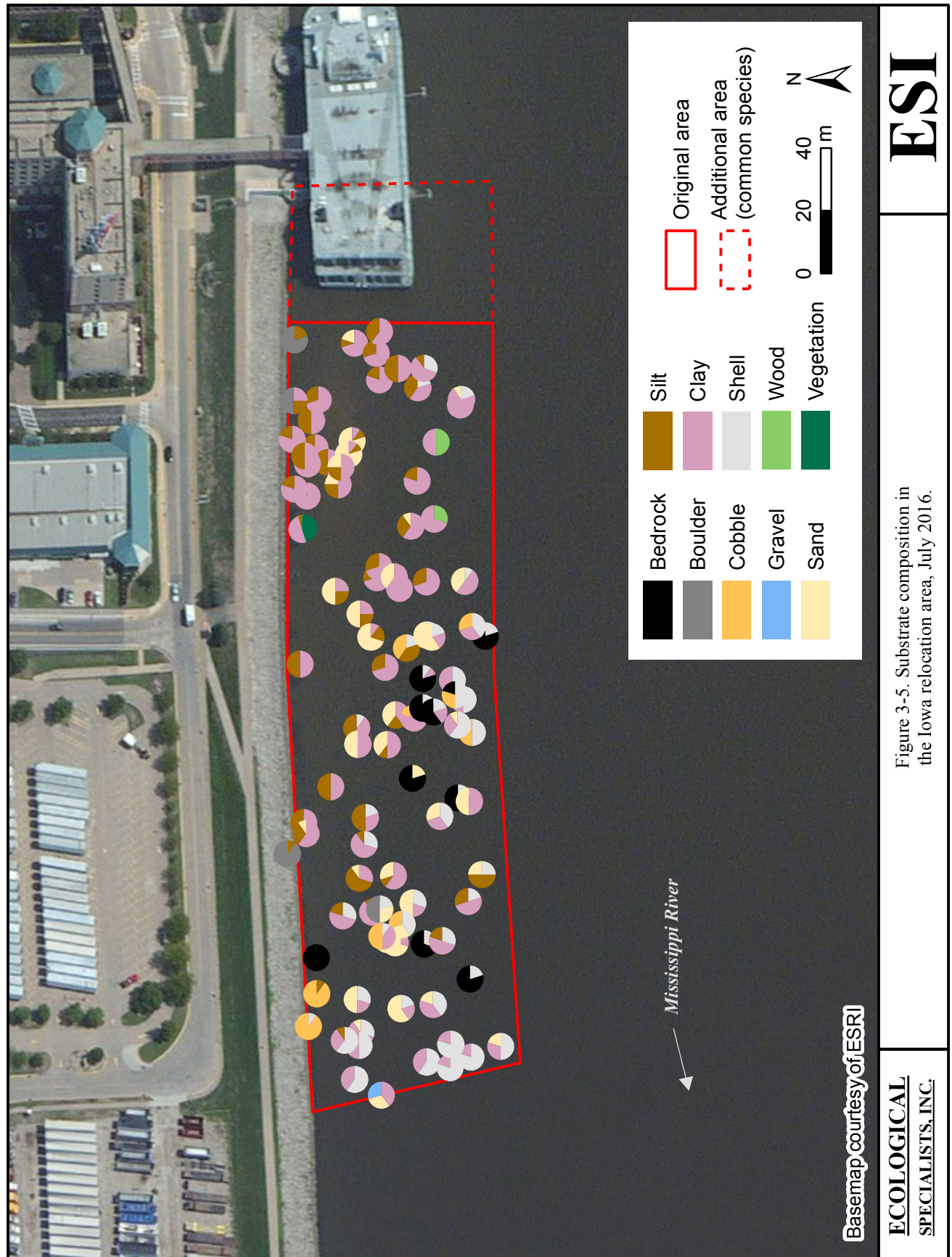


Figure 3-5. Substrate composition in the Iowa relocation area, July 2016.



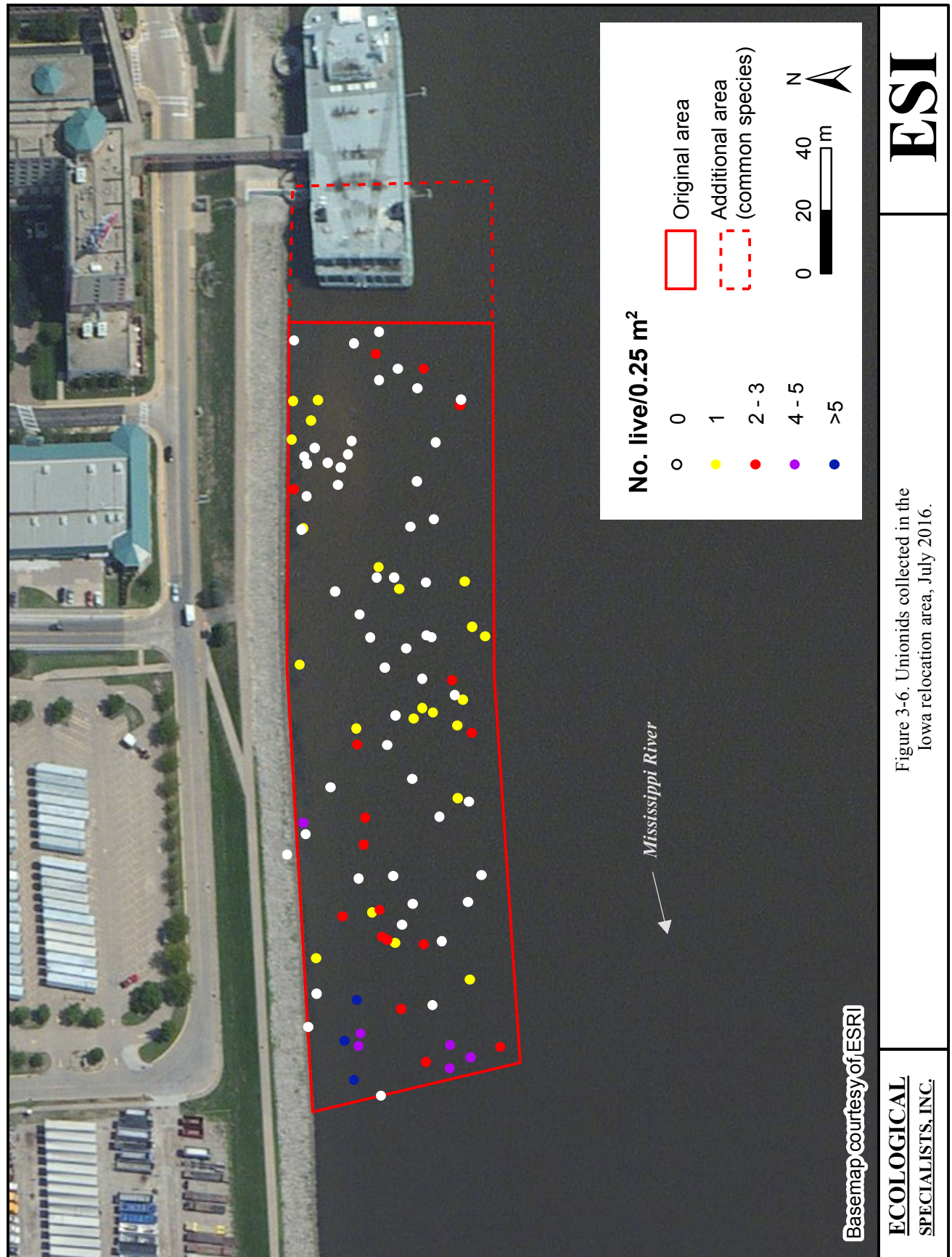


Figure 3-6. Unionids collected in the Iowa relocation area, July 2016.

Table 1-1. Unionid species collected in the I-74 bridge project area, 2014.

Species	Status <sup>1</sup>	IL bank <sup>2</sup>	IA bank <sup>2</sup>
<u>Margaritiferidae</u>			
<i>Cumberlandia monodonta</i>	FE, ILE, IAE	L	L
<u>Amblemini</u>			
<i>Amblema plicata</i>		L	L
<u>Pleurobemini</u>			
<i>Fusconaia ebena</i>	ILT	WD	-
<i>Fusconaia flava</i>		L	L
<i>Plethobasus cyphus</i>	FE, ILE, IAE	L	-
<i>Pleurobema sintoxia</i>	IAE	L	-
<u>Quadrulini</u>			
<i>Cyclonaias tuberculata</i>	ILT, IAT	WD	-
<i>Megalonaias nervosa</i>		L	L
<i>Quadrula metanevra</i>		L	L
<i>Quadrula nodulata</i>		L	L
<i>Quadrula pustulosa</i>		L	L
<i>Quadrula quadrula</i>		L	L
<i>Tritogonia verrucosa</i>	IAE	WD	-
<u>Lampsilini</u>			
<i>Actinonaias ligamentina</i>		WD	-
<i>Ellipsaria lineolata</i>	ILT, IAT	L	L
<i>Lampsilis cardium</i>		L	L
<i>Lampsilis higginsii</i>	FE, ILE, IAE	L	L
<i>Lampsilis teres</i>	IAE	WD	-
<i>Leptodea fragilis</i>		L	L
<i>Ligumia recta</i>	ILT	L	L
<i>Obliquaria reflexa</i>		L	L
<i>Obovaria olivaria</i>		L	L
<i>Potamilus alatus</i>		L	L
<i>Potamilus ohioensis</i>		L	-
<i>Toxolasma parvus</i>		L	-
<i>Truncilla donaciformis</i>		L	L
<i>Truncilla truncata</i>		L	L
<u>Anodontini</u>			
<i>Arcidens confragosus</i>		L	L
<i>Lasmigona complanata</i>		L	L
<i>Pyganodon grandis</i>		L	L
<i>Strophitus undulatus</i>	IAT	WD	-
<i>Utterbackia imbecillis</i>		L	L
Live species		26	22
Total species		32	22

<sup>1</sup> FE = federally endangered, ILE = Illinois endangered, ILT = Illinois threatened, IAE = Iowa endangered, IAT = Iowa threatened. USFWS (2016a), ILDNR (2015), IADNR (2009).

<sup>2</sup> L = live, WD = weathered dead

Table 3-1. Unionids collected in quantitative samples in the Illinois new bridge Action Area, 2014 and 2016.

Species	2016				2014			
	No. Live	%	No. ≤5yo	No. FD	No. Live	%	No. ≤5yo	No. FD
<u>Amblemini</u>								
<i>Amblema plicata</i>	52	15.9	17	7	35	13.6	6	2
<u>Pleurobemini</u>								
<i>Fusconaia ebena</i>	WD	-	-	-	-	-	-	-
<i>Fusconaia flava</i>	7	2.1	2	-	9	3.5	5	-
<u>Quadrulini</u>								
<i>Megaloniaias nervosa</i>	8	2.4	3	2	9	3.5	5	1
<i>Quadrula metanevra</i>	5	1.5	-	-	2	0.8	1	-
<i>Quadrula nodulata</i>	-	-	-	-	WD	-	-	-
<i>Quadrula pustulosa</i>	112	34.3	44	20	95	37.0	30	9
<i>Quadrula quadrula</i>	8	2.4	4	1	4	1.6	-	1
<i>Tritogonia verrucosa</i>	WD	-	-	-	-	-	-	-
<u>Lampsilini</u>								
<i>Ellipsaria lineolata</i>	7	2.1	5	1	4	1.6	2	1
<i>Lampsilis cardium</i>	9	2.8	2	3	7	2.7	2	-
<i>Lampsilis higginsii</i>	1	0.3	-	-	2	0.8	1	-
<i>Leptodea fragilis</i>	7	2.1	6	5	6	2.3	6	5
<i>Ligumia recta</i>	18	5.5	2	-	7	2.7	-	-
<i>Obliquaria reflexa</i>	81	24.8	55	13	59	23.0	36	5
<i>Obovaria olivaria</i>	5	1.5	3	2	2	0.8	-	-
<i>Potamilus alatus</i>	1	0.3	-	-	3	1.2	2	-
<i>Truncilla donaciformis</i>	1	0.3	1	1	4	1.6	4	9
<i>Truncilla truncata</i>	5	1.5	3	-	6	2.3	6	-
<u>Anodontini</u>								
<i>Arcidens confragosus</i>	FD	-	-	1	1	0.4	-	-
<i>Lasmigona complanata</i>	WD	-	-	-	1	0.4	1	-
<i>Pyganodon grandis</i>	WD	-	-	-	-	-	-	-
<i>Utterbackia imbecillis</i>	-	-	-	-	1	0.4	1	15
Total	327	100.0	147	56	257	100.0	108	48
Live species	16				19			
Total species	21				20			
Density (no./m <sup>2</sup> ± 2SE) in whole Action Area <sup>1</sup>	13.1 ± 3.3 <sup>A</sup>				20.6 ± 6.6 <sup>A</sup>			
No. samples	100				50			
Density (no./m <sup>2</sup> ± 2SE) in Piers 1-5	21.8 ± 4.4				31.9 ± 7.8			
No. samples	59				32			
% ≤5 years old	45.0				42.0			
% mortality	14.6				15.7			

FD = fresh dead shell; WD = weathered dead shell

<sup>1</sup> Different letters within a row denote a significant difference (Wilcoxon signed-rank test, p≤0.01)

Table 3-2. Power analysis for the Illinois and Iowa new bridge Action Areas, 2016.

	Illinois new bridge Action Area	Iowa new bridge Action Area
Sample Size	100	50
Mean Density	13.1	12.9
(95% CI)	3.3	3.2
Standard Deviation	16.7	11.2
Precision <sup>1</sup>	25%	25%
Precision level <sup>1</sup>	No. of Samples	
15%	289	134
20%	163	75
25%	104	48

<sup>1</sup>Precision level = 95% CI/mean

Table 3-3. Unionids collected in quantitative samples in the Iowa new bridge Action Area, 2014 and 2016.

Species	2016				2014 <sup>1</sup>			
	No. Live	%	No. ≤5yo	No. FD	No. Live	%	No. ≤5yo	No. FD
<u>Amblemini</u>								
<i>Amblema plicata</i>	17	10.6	-	2	4	44.4	-	-
<u>Pleurobemini</u>								
<i>Fusconaia flava</i>	FD <sup>2</sup>	-	-	1	-	-	-	-
<u>Quadrulini</u>								
<i>Megaloniaias nervosa</i>	12	7.5	-	-	-	-	-	-
<i>Quadrula metanevra</i>	2	1.2	-	-	-	-	-	-
<i>Quadrula nodulata</i>	FD	-	-	1	1	11.1	1	-
<i>Quadrula pustulosa</i>	39	24.2	4	8	1	11.1	1	-
<i>Quadrula quadrula</i>	14	8.7	-	-	1	11.1	-	-
<u>Lampsilini</u>								
<i>Actinoniaias ligamentina</i>	1	0.6	-	-	-	-	-	-
<i>Ellipsaria lineolata</i>	17	10.6	4	-	1	11.1	-	-
<i>Lampsilis cardium</i>	11	6.8	-	-	1	11.1	-	-
<i>Lampsilis higginsii</i>	FD	-	-	1	-	-	-	-
<i>Leptodea fragilis</i>	5	3.1	4	-	-	-	-	-
<i>Ligumia recta</i>	15	9.3	-	1	-	-	-	-
<i>Obliquaria reflexa</i>	22	13.7	7	8	-	-	-	-
<i>Obovaria olivaria</i>	2	1.2	1	-	-	-	-	-
<i>Potamilus alatus</i>	1	0.6	-	1	-	-	-	-
<i>Truncilla truncata</i>	WD	-	-	-	-	-	-	-
<u>Anodontini</u>								
<i>Lasmigona complanata</i>	3	1.9	-	-	-	-	-	-
Total	161	100.0	20	23	9	100.0	2	0
Live species	14				6			
Total species	18				6			
Density (no./m <sup>2</sup> ± 2SE) <sup>3</sup>	12.9 ± 3.2 <sup>A</sup>				3.0 ± 2.7 <sup>B</sup>			
No. samples	50				12			
% ≤5 years old	12.4				22.2			
% mortality	12.5				0.0			

<sup>1</sup> Includes only samples collected shoreward of Pier 2 (within the same area as 2016 samples)

<sup>2</sup> FD = fresh dead shell; WD = weathered dead shell

<sup>3</sup> Different letters within a row denote a significant difference (Wilcoxon signed-rank test, p≤0.01)

Table 3-4. Unionids relocated from the Illinois piers and outfall area, August - October 2016.

Species	Pier 5 + Outfall							Total	%
	Pier 1	Pier 2	Pier 3	Pier 4	Pier 5	Outfall <sup>1</sup>	Outfall		
<u>Margaritiferidae</u>									
<i>Cumberlandia monodonta</i>	15	7	-	-	-	-	1	23	0.017
<u>Amblemini</u>									
<i>Amblema plicata</i>	6,841	5,063	3,336	3,475	2,974	335	201	22,225	15.974
<u>Pleurobemini</u>									
<i>Elliptio dilatata</i>	-	-	1	-	1	-	-	2	0.001
<i>Fusconaia flava</i>	2,194	642	246	189	175	55	40	3,541	2.545
<i>Plethobasus cyphyus</i>	7	18	47	13	21	-	-	106	0.076
<i>Pleurobema sintoxia</i>	6	11	5	5	5	1	-	33	0.024
<u>Quadrulini</u>									
<i>Cyclonaias tuberculata</i>	-	-	1	-	-	-	-	1	0.001
<i>Megaloniais nervosa</i>	1,049	1,171	377	442	444	62	25	3,570	2.566
<i>Quadrula metanevra</i>	679	1,501	681	199	208	6	1	3,275	2.354
<i>Quadrula nodulata</i>	137	33	42	50	15	3	1	281	0.202
<i>Quadrula pustulosa</i>	15,415	11,392	7,884	7,000	7,474	449	278	49,892	35.859
<i>Quadrula quadrula</i>	1,017	353	425	978	835	51	21	3,680	2.645
<u>Lampsilini</u>									
<i>Actinonaias ligamentina</i>	16	16	22	5	21	-	-	80	0.057
<i>Ellipsaria lineolata</i>	676	785	400	484	277	15	3	2,640	1.897
<i>Lampsilis cardium</i>	1,557	1,421	1,710	1,065	801	95	24	6,673	4.796
<i>Lampsilis higginsii</i>	174	177	156	127	113	-	-	747	0.537
<i>Lampsilis siliquoidea</i>	1	-	-	-	-	-	-	1	0.001
<i>Lampsilis teres</i>	1	-	-	2	1	-	-	4	0.003
<i>Leptodea fragilis</i>	225	329	175	157	94	5	1	986	0.709
<i>Ligumia recta</i>	1,641	1,709	2,095	1,755	1,353	130	58	8,741	6.282
<i>Obliquaria reflexa</i>	6,450	7,220	5,681	4,933	4,141	258	99	28,782	20.687
<i>Obovaria olivaria</i>	119	401	403	500	373	13	2	1,811	1.302
<i>Potamilus alatus</i>	209	85	119	74	34	5	6	532	0.382
<i>Potamilus ohioensis</i>	8	2	3	2	2	-	-	17	0.012
<i>Toxolasma parvus</i>	5	2	-	-	-	-	-	7	0.005
<i>Truncilla donaciformis</i>	31	4	9	5	2	-	1	52	0.037
<i>Truncilla truncata</i>	207	89	44	70	15	5	3	433	0.311
<u>Anodontini</u>									
<i>Arcidens confragosus</i>	185	85	66	45	12	9	5	407	0.293
<i>Lasmsgona complanata</i>	304	113	54	56	20	16	3	566	0.407
<i>Pyganodon grandis</i>	15	1	-	2	-	-	1	19	0.014
<i>Strophitus undulatus</i>	2	-	1	1	-	-	-	4	0.003
<i>Utterbackia imbecillis</i>	-	-	2	-	-	-	-	2	0.001
Total	39,186	32,630	23,985	21,634	19,411	1,513	774	139,133	100.000
Live species	29	26	27	26	25	18	20	32	
% ≤5 years old	30.5	20.8	21.1	17.9	6.7	17.8	34.8	21.3	

<sup>1</sup> Unionids from Pier 5 and the outfall were all processed together on 10/24/16. Total from Pier 5 = 20,000; total from outfall = 1,698.



Table 3-5. Expected number of unionids relocated from the Illinois new bridge Action Area.

	2016 data		
	Average	Low	High
<u>Piers 1-5</u>			
Density (no./m <sup>2</sup> )	21.8	17.4	26.2
Total area (all search grids; m <sup>2</sup> )		13,890	
Est. total no. live	302,802	241,686	363,918
Est. no. >25 mm	251,326	200,599	302,052
Est. total relocated	226,193	180,539	271,847
 <u>Outfall</u> <sup>1</sup>			
Density (no./m <sup>2</sup> )	21.8	17.4	26.2
Total area (m <sup>2</sup> )		193	
Est. total no. live	4,207	3,358	5,057
Est. no. >25 mm	3,492	2,787	4,197
Est. total relocated	3,143	2,509	3,777

<sup>1</sup> The outfall area was not sampled in 2016. Data from the new bridge Action Area was extended to the outfall area, as it is only ~15 m downstream of the bridge corridor.

Table 3-6. Age frequency of federal and state T&amp;E species collected from the Illinois piers and outfall area, August - October 2016.

Species	Age (external annuli count)																			Total			
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19		20+	Not recorded	
<u>Federal T&amp;E species</u>																							
<i>Cumberlandia monodonta</i>	-	-	-	-	-	-	1	-	-	-	1	2	1	1	-	1	2	1	1	1	12	-	23
<i>Plethobasus cyphus</i>	-	-	-	1	-	-	-	-	3	4	8	18	15	18	15	10	5	5	1	3	-	-	106
<i>Lampsilis higginsii</i>	-	-	4	15	91	81	60	55	64	57	69	48	46	53	34	26	11	11	13	6	3	-	747
<u>State T&amp;E species</u>																							
<i>Elliptio dilatata</i>	-	-	-	-	-	-	1	-	-	-	1	-	-	-	-	-	-	-	-	-	-	-	2
<i>Cyclonaias tuberculata</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	1
<i>Ellipsaria lineolata</i>	-	-	84	422	685	550	367	240	121	106	49	9	4	-	1	1	-	-	-	-	-	1	2,640
<i>Ligumia recta</i>	2	30	87	191	666	1012	1303	1264	1183	964	776	532	339	201	91	43	31	9	8	8	1	8,741	
Total	2	30	175	629	1442	1643	1732	1559	1371	1131	904	609	405	273	141	81	49	26	24	29	5	12,260	

Table 3-7. Unionids placed in density treatment areas, August - October 2016.

Species	Illiniwek Park		Eagle's Landing		Upstream	
	2x Density	3x Density	2x Density	3x Density	2x Density	3x Density
<u>Amblemini</u>						
<i>Amblema plicata</i>	3,763	4,011	4,061	3,973	2,344	4,073
<u>Pleurobemini</u>						
<i>Fusconaia flava</i>	557	610	614	666	496	598
<i>Pleurobema sintoxia</i>	6	4	4	9	5	5
<u>Quadrulini</u>						
<i>Megaloniaias nervosa</i>	567	675	652	680	401	595
<i>Quadrula metanevra</i>	567	542	574	594	436	562
<i>Quadrula nodulata</i>	68	46	41	41	47	38
<i>Quadrula pustulosa</i>	8,621	8,992	9,031	9,160	5,213	8,875
<i>Quadrula quadrula</i>	691	684	649	672	272	712
<u>Lampsilini</u>						
<i>Actinonaias ligamentina</i>	9	13	9	21	7	21
<i>Ellipsaria lineolata</i>	477	403	453	463	306	400
<i>Lampsilis cardium</i>	1,059	1,159	1,214	1,249	740	1,253
<i>Lampsilis siliquoidea</i>	-	1	-	-	-	-
<i>Lampsilis teres</i>	-	1	-	-	-	-
<i>Leptodea fragilis</i>	165	171	179	175	116	180
<i>Ligumia recta</i>	1,516	1,559	1,611	1,544	829	1,682
<i>Obliquaria reflexa</i>	4,950	5,205	5,299	5,350	2,898	5,080
<i>Obovaria olivaria</i>	331	374	336	327	115	328
<i>Potamilus alatus</i>	104	84	97	85	68	94
<i>Potamilus ohiensis</i>	2	6	4	2	-	3
<i>Toxolasma parvus</i>	3	-	1	2	-	1
<i>Truncilla donaciformis</i>	4	10	12	9	9	8
<i>Truncilla truncata</i>	74	100	84	64	32	79
<u>Anodontini</u>						
<i>Arcidens confragosus</i>	68	90	71	67	53	58
<i>Lasmigona complanata</i>	91	91	120	96	68	100
<i>Pyganodon grandis</i>	1	3	6	2	2	5
<i>Strophitus undulatus</i>	-	2	2	-	-	-
<i>Utterbackia imbecillis</i>	-	1	-	1	-	-
Total	23,694	24,837	25,124	25,252	14,457	24,750

Table 3-8. Unionids placed in endangered species grids, August - October 2016.

Species	Sylvan Slough <i>C. monodonta</i>	Illiniwek Park <i>L. higginsii</i>	Illiniwek Park <i>P. cyphyus</i>	Illiniwek Park <i>P. cyphyus</i>	Eagle's Landing <i>L. higginsii</i>	Eagle's Landing <i>P. cyphyus</i>	Upstream L. <i>higginsii</i>	USFWS - Genoa	Total
<u>Federal T&amp;E species</u>									
<i>Cumberlandia monodonta</i>	24	-	-	-	-	-	-	-	24
<i>Plethobasus cyphus</i>	-	-	50	-	-	57	-	-	107
<i>Lampsilis higginsii</i>	-	227	-	-	225	-	242	62	756 <sup>1</sup>
<u>Other species</u>									
<i>Elliptio dilatata</i>	-	-	1	1	-	1	-	-	2
<i>Cyclonaias tuberculata</i>	-	-	1	-	-	-	-	-	1
<i>Ellipsaria lineolata</i>	-	-	-	-	-	-	-	138	138
<i>Lampsilis teres</i>	-	-	3	-	-	-	-	-	3 <sup>2</sup>
Total	24	227	55	55	225	58	242	200	831

<sup>1</sup> Includes 1 marked *L. higginsii* collected in the Illiniwek Park 2x Density area (see Table 3-11).

<sup>2</sup> The fourth *L. teres* collected in the relocation was randomly assigned to the Illiniwek Park 3x Density area (see Table 3-5).

Table 3-9. Unionids relocated from the Iowa piers, October 2016.

Species	Pier 1	Pier 2	Pier 3	Total	%
<u>Margaritiferidae</u>					
<i>Cumberlandia monodonta</i>	-	1	-	1	0.06
<u>Amblemini</u>					
<i>Amblema plicata</i>	155	52	31	238	15.25
<u>Pleurobemini</u>					
<i>Fusconaia flava</i>	5	-	-	5	0.32
<i>Plethobasus cyphus</i>	1	-	-	1	0.06
<u>Quadrulini</u>					
<i>Megaloniaias nervosa</i>	178	28	10	216	13.84
<i>Quadrula metanevra</i>	24	10	8	42	2.69
<i>Quadrula pustulosa</i>	175	84	13	272	17.42
<i>Quadrula quadrula</i>	129	12	4	145	9.29
<u>Lampsilini</u>					
<i>Actinonaias ligamentina</i>	1	-	-	1	0.06
<i>Ellipsaria lineolata</i>	74	15	11	100	6.41
<i>Lampsilis cardium</i>	22	19	1	42	2.69
<i>Lampsilis higginsii</i>	5	1	2	8	0.51
<i>Leptodea fragilis</i>	13	-	2	15	0.96
<i>Ligumia recta</i>	74	44	31	149	9.55
<i>Obliquaria reflexa</i>	102	123	13	238	15.25
<i>Obovaria olivaria</i>	20	20	10	50	3.20
<i>Potamilus alatus</i>	2	1	-	3	0.19
<i>Truncilla donaciformis</i>	3	-	-	3	0.19
<i>Truncilla truncata</i>	3	1	-	4	0.26
<u>Anodontini</u>					
<i>Arcidens confragosus</i>	28	-	-	28	1.79
Total	1,014	411	136	1,561	100.00
Live species	19	14	12	20	
% ≤5 years old	6.4	4.6	4.4	5.8	

Table 3-10. Expected number of unionids relocated from the Iowa new bridge Action Area.

	2016 data		
	Average	Low	High
<u>Pier 1</u>			
Density (no./m <sup>2</sup> )	12.9	9.7	16.1
Total area of search grid (m <sup>2</sup> )		1,570	
Est. total no. live	20,253	15,229	25,277
Est. no. >25 mm	20,050	15,077	25,024
Est. total relocated	18,045	13,569	22,522
<u>Pier 2</u>			
Density (no./m <sup>2</sup> )	12.9	9.7	16.1
Total area of search grid (m <sup>2</sup> )		1,539	
Est. total no. live	19,853	14,928	24,778
Est. no. >25 mm	19,655	14,779	24,530
Est. total relocated	17,689	13,301	22,077

Table 3-11. Age frequency of federal and state T&amp;E species collected from the Iowa piers, October 2016.

Species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20+	Total	
<u>Federal T&amp;E species</u>																						
<i>Cumberlandia monodonta</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	1
<i>Plethobasus cyphus</i>	-	-	-	-	-	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1
<i>Lampsilis higginsii</i>	-	-	-	-	-	-	1	-	2	2	-	-	-	-	-	-	1	2	-	-	-	8
<u>State T&amp;E species</u>																						
<i>Ellipsaria lineolata</i>	-	-	-	3	13	17	30	18	9	4	1	1	-	3	-	-	1	-	-	-	-	100
Total	-	-	-	3	13	18	31	18	11	6	1	1	0	3	0	0	2	2	0	1	1	110

Table 3-12. Unionids collected in the Iowa relocation area, July 2016.

Species	No. Live	%	No. ≤5yo	No. FD
<u>Amblemini</u>				
<i>Amblema plicata</i>	14	12.3	2	-
<u>Pleurobemini</u>				
<i>Fusconaia flava</i>	WD	-	-	-
<u>Quadrulini</u>				
<i>Megaloniaias nervosa</i>	7	6.1	-	-
<i>Quadrula nodulata</i>	WD	-	-	-
<i>Quadrula pustulosa</i>	15	13.2	4	-
<i>Quadrula quadrula</i>	16	14.0	3	-
<u>Lampsilini</u>				
<i>Actinonaias ligamentina</i>	1	0.9	1	-
<i>Ellipsaria lineolata</i>	3	2.6	1	-
<i>Lampsilis cardium</i>	2	1.8	-	-
<i>Lampsilis higginsii</i>	FD	-	-	1
<i>Leptodea fragilis</i>	25	21.9	25	1
<i>Ligumia recta</i>	WD	-	-	-
<i>Obliquaria reflexa</i>	21	18.4	16	1
<i>Obovaria olivaria</i>	2	1.8	-	-
<i>Potamilus alatus</i>	WD	-	-	-
<i>Truncilla donaciformis</i>	2	1.8	2	-
<i>Truncilla truncata</i>	3	2.6	3	-
<u>Anodontini</u>				
<i>Arcidens confragosus</i>	1	0.9	-	-
<i>Lasmigona complanata</i>	1	0.9	-	-
<i>Pyganodon grandis</i>	WD	-	-	-
<i>Utterbackia imbecillis</i>	1	0.9	1	-
Total	114	100.0	58	3
Live species	15			
Total species	21			
Density (no./m <sup>2</sup> ± 2SE)	4.6 ± 1.4			
No. samples	100			
% ≤5 years old	50.9			
% mortality	2.6			
Area (m <sup>2</sup> )	16,119			
No. unionids that could be placed at 50% density increase	37,074			

FD = fresh dead shell; WD = weathered dead shell



Table 3-13. Habitat characteristics in density treatment areas, July and November 2016.

Species	Depth (m)		Average % substrate constituents												
	Min.	Max.	Bedrock	Boulder	Cobble	Gravel	Sand	Silt	Clay	Shell	Detritus	Wood	Vegetation		
<u>Illiniwek Park</u> Control area	0.9	1.5	0.0	0.0	0.0	0.0	0.1	27.1	72.9	0.0	0.0	0.0	0.0		
2x Density - pre-relo	1.8	3.4	0.0	0.3	0.6	0.5	31.4	31.5	8.6	25.9	0.0	1.3	0.0		
2x Density - post-relo	2.4	3.7	0.8	0.0	0.6	0.8	59.8	11.3	3.0	23.9	0.0	0.0	0.0		
3x Density - pre-relo	1.2	2.4	0.0	0.0	0.0	0.0	0.1	26.8	72.8	0.1	0.0	0.2	0.0		
3x Density - post-relo	1.2	1.8	0.0	0.0	0.0	0.0	0.0	46.3	47.3	6.5	0.0	0.0	0.0		
<u>Eagle's Landing</u> Control area	0.3	2.7	0.0	0.0	18.3	2.9	20.4	21.2	0.4	36.9	0.0	0.0	0.0		
2x Density - pre-relo	0.3	2.7	0.0	0.0	1.4	0.0	1.9	58.4	0.0	38.3	0.0	0.0	0.0		
2x Density - post-relo	1.2	3.7	0.0	0.0	2.0	0.0	44.5	17.3	16.8	19.3	0.3	0.0	0.0		
3x Density - pre-relo	0.6	2.1	0.0	0.3	7.5	0.3	24.5	21.5	8.4	37.2	0.0	0.4	0.0		
3x Density - post-relo	1.5	2.7	0.0	0.0	1.5	4.3	34.5	9.5	16.0	28.5	5.8	0.0	0.0		
<u>Upstream</u> Control area	0.3	3.4	1.8	0.0	1.6	1.9	19.9	34.4	1.5	37.0	0.0	1.9	0.0		
2x Density - pre-relo	1.5	4.0	3.8	0.9	0.6	0.0	30.3	33.5	6.9	21.6	0.0	2.5	0.0		
2x Density - post-relo	2.7	4.0	2.5	0.0	0.8	3.3	63.5	8.5	0.8	20.5	0.3	0.0	0.0		
3x Density - pre-relo	0.6	3.0	6.1	0.1	0.7	0.0	11.7	43.8	17.4	19.6	0.0	0.5	0.1		
3x Density - post-relo	1.2	3.7	3.5	0.0	1.0	1.0	21.3	29.0	18.5	24.8	1.0	0.0	0.0		

Table 3-14. Unionids collected in Illiniwek Park treatment areas before and after placement of relocated unionids.

Species	Control	2x Density			3x Density		
	Pre-relo	Pre-relo	Post-relo		Pre-relo	Post-relo	
			Resident	Relocated		Resident	Relocated
<u>Amblemini</u>							
<i>Amblema plicata</i>	109	93	33	13	235	43	14
<u>Pleurobemini</u>							
<i>Fusconaia flava</i>	16	11	3	1	18	4	1
<u>Quadrulini</u>							
<i>Megaloniaias nervosa</i>	-	5	4	3	-	-	5
<i>Quadrula metanevra</i>	-	1	-	-	-	1	2
<i>Quadrula nodulata</i>	1	2	-	-	1	-	-
<i>Quadrula pustulosa</i>	4	109	25	31	27	9	40
<i>Quadrula quadrula</i>	4	4	1	6	6	2	8
<u>Lampsilini</u>							
<i>Actinonaias ligamentina</i>	-	1	1	-	-	-	-
<i>Ellipsaria lineolata</i>	-	20	3	2	1	-	1
<i>Lampsilis cardium</i>	-	7	7	5	-	2	6
<i>Lampsilis higginsii</i>	-	1	-	1	-	-	-
<i>Lampsilis siliquoidea</i>	-	1	-	-	-	-	-
<i>Lampsilis teres</i>	1	-	-	-	-	-	-
<i>Leptodea fragilis</i>	1	37	5	-	27	3	-
<i>Ligumia recta</i>	-	7	4	13	-	-	4
<i>Obliquaria reflexa</i>	68	85	32	15	211	42	15
<i>Obovaria olivaria</i>	-	5	2	3	-	-	2
<i>Potamilus alatus</i>	3	-	-	-	8	3	-
<i>Potamilus ohioensis</i>	-	-	-	-	1	-	-
<i>Toxolasma parvus</i>	2	-	-	-	5	1	-
<i>Truncilla donaciformis</i>	2	2	4	-	19	6	-
<i>Truncilla truncata</i>	1	8	1	1	2	-	-
<u>Anodontini</u>							
<i>Lasmigona complanata</i>	1	-	-	-	3	1	2
<i>Pyganodon grandis</i>	2	-	-	-	2	-	-
<i>Utterbackia imbecillis</i>	-	-	2	-	10	-	-
Total	215	399	127	94	576	117	100
Live species	14	18	15	12	16	12	12
Density (no./m <sup>2</sup> ± 2SE)	10.8 ± 1.9	20.0 ± 2.5	12.7 ± 3.4	9.4 ± 4.2	28.8 ± 4.9	12.0 ± 3.3	10.3 ± 5.3
% ≤5 years old	57.7	45.4	52.0	18.1	53.0	59.8	16.0
% mortality	2.3	0.7	19.1	0.0	2.2	9.3	2.9

Table 3-15. Comparison of pre- and post-relocation unionid density in relocation areas.

	Illiniwek Park		Eagle's Landing		Upstream	
	2x Density	3x Density	2x Density	3x Density	2x Density	3x Density
Pre-relocation density (no./m <sup>2</sup> ± 2SE)	20.0 ± 2.5	28.8 ± 4.9	61.5 ± 10.2	91.8 ± 19.8	11.8 ± 2.2	11.1 ± 2.4
Adjusted density (factoring out young/small individuals)	18.5	22.4	47.8	69.1	10.9	10.0
Estimated initial population of area	23,791	28,806	61,471	88,863	14,017	12,860
Additional unionids needed to double/triple density	23,791	57,613	61,471	177,725	14,017	25,720
Actual no. unionids placed	23,694	24,837	25,124	25,252	14,457	24,750
Post-relocation density of resident unionids (no./m <sup>2</sup> ± 2SE)	12.7 ± 3.4	12.0 ± 3.3	24.2 ± 5.0	36.1 ± 8.7	8.8 ± 2.4	10.8 ± 3.4
Post-relocation density of relocated unionids (no./m <sup>2</sup> ± 2SE)	9.4 ± 4.2	10.3 ± 5.3	7.6 ± 3.6	6.2 ± 3.9	4.7 ± 1.9	8.1 ± 3.2
Expected density of relocated unionids (no./m <sup>2</sup> ) <sup>1</sup>	18.4	19.3	19.5	19.6	11.2	19.2
Target % change in density	100.0	200.0	100.0	200.0	100.0	200.0
Expected % change in density	99.6	86.2	40.9	28.4	103.1	192.5
Observed % change in density <sup>2</sup>	74.0	85.8	31.4	17.2	53.4	75.0
Wilcoxon test comparing pre- and post-relocation resident densities (p value)	0.003	0.001	0.000	0.000	0.077	1.000
Wilcoxon test comparing post-relocation resident and relocated unionid densities (p value)	0.136	0.259	0.000	0.000	0.017	0.110

<sup>1</sup> Actual no. unionids placed divided by treatment area size (1,286 m<sup>2</sup>)

<sup>2</sup> Uses post-relocation data only; density of relocated unionids divided by density of resident unionids

Table 3-16. Unionids collected in Eagle's Landing treatment areas before and after placement of relocated unionids.

Species	Control		2x Density		3x Density		
	Pre-relo	Pre-relo	Post-relo		Pre-relo	Post-relo	
			Resident	Relocated		Resident	Relocated
<u>Amblemini</u>							
<i>Amblema plicata</i>	122	133	29	12	217	56	14
<u>Pleurobemini</u>							
<i>Fusconaia flava</i>	20	4	4	-	31	10	-
<u>Quadrulini</u>							
<i>Megaloniaias nervosa</i>	93	13	1	2	69	32	4
<i>Quadrula metanevra</i>	8	5	1	1	14	3	2
<i>Quadrula nodulata</i>	1	2	-	-	1	-	-
<i>Quadrula pustulosa</i>	205	155	39	21	276	58	13
<i>Quadrula quadrula</i>	107	70	22	4	111	50	7
<u>Lampsilini</u>							
<i>Actinonaias ligamentina</i>	-	-	-	1	-	-	-
<i>Ellipsaria lineolata</i>	55	46	22	2	71	14	3
<i>Lampsilis cardium</i>	5	4	4	3	8	3	2
<i>Lampsilis higginsii</i>	-	2	-	-	1	-	-
<i>Leptodea fragilis</i>	136	41	14	-	165	21	-
<i>Ligumia recta</i>	7	3	1	10	6	4	6
<i>Obliquaria reflexa</i>	194	363	66	16	292	52	8
<i>Obovaria olivaria</i>	7	14	5	1	16	1	2
<i>Potamilus alatus</i>	16	23	7	1	33	7	1
<i>Potamilus ohiensis</i>	1	-	-	-	1	-	-
<i>Toxolasma parvus</i>	2	-	-	-	9	4	-
<i>Truncilla donaciformis</i>	98	91	9	-	76	17	-
<i>Truncilla truncata</i>	110	60	16	-	91	18	-
<u>Anodontini</u>							
<i>Arcidens confragosus</i>	1	-	2	1	6	6	-
<i>Lasmigona complanata</i>	3	1	-	1	3	2	-
<i>Pyganodon grandis</i>	-	1	-	-	1	-	-
<i>Strophitus undulatus</i>	-	-	-	-	1	-	-
<i>Utterbackia imbecillis</i>	368	198	-	-	336	3	-
Total	1,559	1,229	242	76	1,835	361	62
Live species	21	20	16	14	24	19	11
Density (no./m <sup>2</sup> ± 2SE)	78.0 ± 15.9	61.5 ± 10.2	24.2 ± 5.0	7.6 ± 3.6	91.8 ± 19.8	36.1 ± 8.7	6.2 ± 3.9
% ≤5 years old	78.6	75.7	66.1	15.8	75.5	61.8	16.1
% mortality	4.5	5.8	14.2	5.0	4.0	8.4	3.1

Table 3-17. Unionids collected in Upstream treatment areas before and after placement of relocated unionids.

Species	Control		2x Density		3x Density		
	Pre-relo	Pre-relo	Post-relo		Pre-relo	Post-relo	
			Resident	Relocated		Resident	Relocated
<u>Amblemini</u>							
<i>Amblema plicata</i>	85	46	13	8	33	17	23
<u>Pleurobemini</u>							
<i>Fusconaia flava</i>	-	3	5	-	4	-	4
<i>Pleurobema sintoxia</i>	-	-	-	1	-	-	-
<u>Quadrulini</u>							
<i>Megaloniaias nervosa</i>	3	5	-	-	4	2	3
<i>Quadrula metanevra</i>	2	-	1	4	-	-	-
<i>Quadrula nodulata</i>	-	-	-	1	-	-	-
<i>Quadrula pustulosa</i>	81	63	26	19	42	28	16
<i>Quadrula quadrula</i>	8	13	5	-	3	-	5
<u>Lampsilini</u>							
<i>Ellipsaria lineolata</i>	19	13	1	1	16	5	1
<i>Lampsilis cardium</i>	14	2	4	1	2	7	5
<i>Lampsilis higginsii</i>	1	-	-	-	-	-	-
<i>Leptodea fragilis</i>	17	19	3	-	14	2	-
<i>Ligumia recta</i>	16	3	1	6	3	4	11
<i>Obliquaria reflexa</i>	90	58	19	5	81	30	10
<i>Obovaria olivaria</i>	5	2	3	-	3	1	2
<i>Potamilus alatus</i>	2	5	1	1	4	4	-
<i>Toxolasma parvus</i>	-	-	-	-	-	2	-
<i>Truncilla donaciformis</i>	2	1	1	-	-	1	-
<i>Truncilla truncata</i>	8	-	4	-	9	3	-
<u>Anodontini</u>							
<i>Arcidens confragosus</i>	-	1	1	-	1	-	-
<i>Lasmigona complanata</i>	2	1	-	-	2	2	1
Total	355	235	88	47	221	108	81
Live species	16	15	15	10	15	14	11
Density (no./m <sup>2</sup> ± 2SE)	17.8 ± 2.6	11.8 ± 2.2	8.8 ± 2.4	4.7 ± 1.9	11.1 ± 2.4	10.8 ± 3.4	8.1 ± 3.2
% ≤5 years old	52.1	40.0	73.9	23.4	59.3	46.3	8.6
% mortality	13.2	17.0	24.1	4.1	31.8	35.7	6.9

Appendix A.

ID numbers of unionids placed in endangered species grids, August – October 2016.

Appendix A-1. Individuals placed in the Illiniwek Park *Lampsilis higginsii* grid, August - October 2016.

Species	Cell 1	Cell 2	Cell 3	Cell 4	
<i>Lampsilis higginsii</i>	16-008 16-009 16-017 16-057 16-066 16-093 16-109 16-117 16-118 16-138 16-171 16-173 16-183 16-188 16-189 16-215 16-229 16-260 16-280 16-284 16-292 16-320 16-349 16-360 16-362 16-369	16-004 16-011 16-016 16-027 16-034 16-040 16-043 16-065 16-075 16-086 16-110 16-124 16-178 16-182 16-194 16-206 16-221 16-223 16-226 16-227 16-232 16-245 16-250 16-251 16-254 16-255 16-281 16-287 16-294 16-313 16-314	16-340 16-356 16-377 16-403 16-411 16-416 16-454 16-458 16-464 16-465 16-467 16-479 16-480 16-482 16-487 16-489 16-491 16-494 16-544 16-587 16-588 16-594 16-613 16-632 16-642 16-652 16-653 16-680 16-733 16-733(a)	16-002 16-015 16-038 16-041 16-050 16-064 16-083 16-084 16-089 16-096 16-106 16-113 16-137 16-160 16-167 16-170 16-177 16-180 16-207 16-214 16-220 16-230 16-247 16-262 16-317 16-318 16-363 16-364	16-030 16-036 16-076 16-077 16-094 16-108 16-112 16-127 16-140 16-149 16-150 16-153 16-169 16-181 16-192 16-205 16-224 16-225 16-228 16-234 16-237 16-241 16-270 16-277 16-285 16-310 16-330 16-336 16-378 16-388
Total no. placed in cell	52	61	55	59	

Appendix A-2. Individuals placed in the Illiniwek Park *Plethobasus cyphus* grid, August - October 2016.

Species	Cell 1	Cell 2	Cell 3	Cell 4
<i>Plethobasus cyphus</i>	16-001	16-040	16-009	16-029
	16-002	16-044	16-027	16-030
	16-003	16-053	16-028	16-039
	16-004	16-054	16-043	16-049
	16-005	16-059	16-078	16-058
	16-013	16-070	16-079	16-060
	16-016	16-071	16-091	16-064
	16-023	16-073		16-072
	16-026	16-074		16-080
	16-032	16-088		
	16-034	16-093		
<i>Elleptio dilatata</i>	-	213D	-	-
<i>Cyclonaias tuberculata</i>	-	2175	-	-
<i>Lampsilis teres</i>	-	-	3 live (no IDs)	-
Total no. placed in cell	22	14	10	9



Appendix A-3. Individuals placed in the Eagle's Landing *Lampsilis higginsii* grid, August - October 2016.

Species	Cell 1	Cell 2	Cell 3	Cell 4	Old grid					
<i>Lampsilis higginsii</i>	16-010	16-327	16-081	16-347	16-073A	16-384	16-088	16-471	16-005	
	16-026	16-332	16-082	16-353	16-085	16-392	16-099	16-481	16-007	
	16-100	16-333	16-092	16-417	16-130	16-405	16-175	16-493	16-012	
	16-114	16-337	16-095	16-429	16-131	16-436	16-179	16-498	16-013	
	16-121	16-352	16-104	16-430	16-161	16-452	16-203	16-505	16-014	
	16-133	16-399	16-116	16-444	16-168	16-466	16-204	16-513	16-018	
	16-139	16-407	16-143	16-445	16-219	16-469	16-211	16-515	16-020	
	16-145	16-420	16-147	16-453	16-248	16-478	16-216	16-522	16-021	
	16-156	16-422	16-159	16-455	16-261	16-502	16-246	16-536	16-023	
	16-172	16-427	16-163	16-462	16-267	16-504	16-257	16-552	16-025	
	16-187	16-450	16-185	16-472	16-269	16-506	16-282	16-565	16-028	
	16-197	16-451	16-201	16-573	16-271	16-511	16-293	16-567	16-032	
	16-200	16-459	16-202	16-579	16-283	16-521	16-300	16-592	16-033	
	16-218	16-485	16-208	16-593	16-305	16-548	16-355	16-603	IL-01	
	16-235	16-490	16-239	16-601	16-309	16-550	16-358	16-605	16-037	
	16-243	16-495	16-264	16-634	16-319	16-596	16-359	16-606	16-039	
	16-258	16-514	16-272	16-645	16-324	16-628	16-361	16-615	16-042	
	16-259	16-530	16-279	16-658	16-338	16-644	16-367	16-617	16-045	
	16-263	16-591	16-289	16-665	16-344	16-659	16-390	16-621	16-046	
	16-288	16-592	16-296	16-715	16-345	16-674	16-393	16-629	16-048	
	16-291	16-608	16-298	16-718	16-354	16-679	16-398	16-639	16-053	
	16-295	16-633	16-299	16-725	16-370	16-699	16-421	16-646	16-056	
	16-297	16-641	16-329	16-729	16-380	16-700	16-424	16-650	16-061	
	16-302	16-649					16-434	16-682	16-062	
	16-306	16-686					16-441	16-698	16-068	
	16-323	16-717					16-449	16-728	16-070	
							16-456	16-732	16-072	
	Total	52	46	46	54	27				

Appendix A-4. Individuals placed in the Eagle's Landing *Plethobasus cyphus* grid, August - October 2016.

Species	Cell 1	Cell 2	Cell 3	Cell 4
<i>Plethobasus cyphus</i>	16-011	16-008	16-014	16-007
	16-015	16-010	16-019	16-012
	16-046	16-017	16-025	16-018
	16-063	16-020	16-031	16-022
	16-081	16-051	16-038	16-024
	16-083	16-061	16-042	16-035
	16-090	16-069	16-047	16-036
	16-096	16-076	16-055	16-041
	16-100	16-087	16-056	16-041
	16-103	16-092	16-075	16-050
				16-052
				16-057
				16-062
				16-065
			16-066	
			16-089	
			16-098	
			16-101	
			16-105	
<i>Elliptio dilatata</i>	-	-	217E	-
Total	10	10	20	18

