

City of Lockport, Illinois

Incidental Take Authorization Application

Conservation Plan for Banded Killifish (*Fundulus diaphanus*)

IL 7 Frontage Road Bridge over the Des Plaines River

Structure No. 099-0135



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LIST OF ABBREVIATIONS

FEMA	-	Federal Emergency Management Agency
IDNR	-	Illinois Department of Natural Resources
IDOT	-	Illinois Department of Transportation
IEPA	-	Illinois Environmental Protection Agency
MWRD		Metropolitan Water Reclamation District of Greater Chicago
USACE	-	United States Army Corps of Engineers
USFWS	-	United States Fish and Wildlife Service

1. INTRODUCTION

1.1 Project Description

The City of Lockport is planning to rehabilitate the IL 7 Frontage Road Bridge that crosses over the Des Plaines River. To complete this action, an Incidental Take Authorization is needed from the Illinois Department of Natural Resources (IDNR) for construction activities that may potentially take banded killifish (*Fundulus diaphanus*) individuals.

The IL 7 Frontage Road Bridge (Structure No. 099-0135) is located over the Des Plaines River immediately north of the IL 7 Bridge in Lockport, Illinois (Exhibit A). The bridge is located in the northeast quarter of Section 22, Township 36 North, Range 10 East in Will County (Latitude 41.592406, Longitude 88.068779). The City of Lockport maintains and has jurisdiction of the IL Route 7 Frontage Road Bridge. Ownership of the action area includes City of Lockport right-of-way, Metropolitan Water Reclamation District of Greater Chicago (MWRD) property, and an unclaimed parcel of land. Work within MWRD property will be completed by permit. Control of the unclaimed parcel is being actively sought.

The two-lane bridge was constructed in 1957 and is currently rated as Structurally Deficient and Functionally Obsolete. It is currently closed to all traffic.

The bridge is an eight-span steel stringer structure with a cast-in-place concrete deck. The seven piers and abutments have spread footings founded on rock.

The proposed rehabilitation includes structural steel repairs to return the bridge to HS-20 capacity, cleaning and painting the structural steel, deck overlay, approach slab replacement, crash railing installation, approach guardrail and terminal replacement. The existing piers will remain.

Since the bridge cannot safely support any traffic, including any potential construction equipment, temporary causeways will need to be constructed from both banks of the Des Plaines River to access Piers 2 and 6 to allow construction equipment to access deteriorated structural steel (Exhibit B). The temporary causeways will be constructed from clean rock sources meeting Illinois Environmental Protection Agency (IEPA)/U.S. Army Corps of Engineers (USACE) requirements for temporary cofferdams in rivers.

1.2 Biological Data

The banded killifish (*Fundulus diaphanus*) is listed as threatened under the Illinois Endangered Species Act (IDNR, 2017a). Several occurrences of the fish have been reported in the lower Des Plaines River (IDNR, 2017a & 2017b; Pescitelli, 2015).

In Illinois, the banded killifish is typically found in clear, glacial lakes (IDNR, 2017c). It can also be found in streams and rivers. Rivera et al. (2013) found the killifish in Mill Creek in Rock Island

County. Based on museum specimens, Tiemann et al. (2015) reported the fish in Coon Creek (Whiteside County), Pools 19 and 20 of the Mississippi River (Hancock County), and Sandy Creek (Putnam County). It has also been reported in the Des Plaines River near the project site (IDNR, 2017b; Pescitelli, 2015) and the Cal-Sag Canal, Illinois River system, and Chicago River system (MWH and J. Meldrim, 2016).

Fish surveys between the IL 7 Bridge and Division Street Bridge in Lockport, just downstream of the IL 7 Frontage Road Bridge, have collected the banded killifish numerous times between 2012 and 2016 (IDNR, 2017b). The number of banded killifish collected from each survey ranged between 0 and 161 with most survey events between 0 and 2 fish. The river habitat at the fish survey areas is similar to the project site, namely an island complex habitat with rock/cobble substrate. Due to the high gradient, the velocity of the flow is relatively rapid.

Throughout its range, the banded killifish has been found in glacial lakes as well as quiet backwaters and sluggish reaches of medium to large streams with low gradients (IDNR, 2017c; MWH and J. Meldrim, 2016). At the project site, the fish likely prefers the downstream side of islands as well as slower flow areas along the river banks. The schools of the banded killifish are typically found over sand, gravel or detritus-covered bottom with patches of submerged aquatic plants (NatureServe, 2017). Reports of macrophytic vegetation vary from none to dense (WDNR, 2017 and UM, 2017)

Spawning occurs in late spring and early summer (IDNR, 2017c). The eggs are deposited in small clusters that stick to aquatic plants. Larval banded killifish move towards the shore and are found singly or in small numbers (fishbabies.ca, 2017).

The banded killifish is non-migratory (NatureServe, 2017; Rey and Turgeon, 2007). It is found in small schools. During the day, the fish are found in shallow water but move into deeper water at night (COSEWIC, 2014).

They eat a variety of aquatic invertebrates (IDNR, 2017c; Rivera et al., 2013). It feeds equally mid-water as well as the bottom and surface (Smith, 2002).

The diversity of fish species is improving in the lower Des Plaines River (Pescitelli, 2015). More sport fishes, intolerant-to-pollution species, and specialist species are occurring, probably as a result of improved water quality conditions.

The number and diversity of mussels is also improving. Threatened or endangered mussels are not likely to occur at the project site (N. Grider, pers. comm., 2017; Pescitelli, 2015). The nearest quality mussel bed is located near the confluence of the Des Plaines River, Kankakee River, and Illinois River. The Brandon Dam, which minimizes fish (and mussel) migration, is located between the mussel bed and the project site.

1.3 Description of the Activities That May Result in Taking

Temporary causeways are needed to access the piers and bridge components requiring rehabilitation (Exhibit B). The temporary causeways will be constructed from clean rock sources meeting Illinois Environmental Protection Agency (IEPA)/U.S. Army Corps of Engineers (USACE) requirements for temporary cofferdams in rivers. The causeways will be approximately 30 feet wide and will extend approximately 0.5 foot above the surface water level. Culverts will be placed in the causeways to allow water to flow through them.

The temporary rock will be brought to the site in trucks and dumped into piles in the upland area. Construction of the causeways will proceed from the shoreline with front-end loaders and track-mounted excavators and dozers moving the rock from the upland stockpiles and depositing and grading it to form the causeway. Once the causeway is no longer needed, the causeway will be removed in reverse order – starting at the riverward end of the causeway, excavators will remove the rock from the river and dozers and front-end loaders will move the rock back to the upland areas for subsequent loading in trucks and removal from the site.

To minimize impacts to habitat, only one causeway will be constructed and operated at a time. Once the construction work is completed for the first causeway, then it will be removed and the second causeway will be built. This leaves at least one side of the river available to the banded killifish at all times.

1.4 Anticipated Adverse Effects on Listed Species

The causeways will temporarily occupy potential banded killifish habitat. It is possible the placement of rock could directly strike a banded killifish, but the construction noise will likely keep any fish away from the causeway construction zone.

Bridge construction noise and vibrations could also deter the fish from occupying potential habitat at the project site. The noise and vibrations against the steel structures and piers could permeate into the water column, which fish would naturally avoid and swim away.

At construction sites, soil particles can enter the water column due to erosive conditions and poor soil erosion and sediment control. Sediment can cover the rock substrate. Suspended soil particles can make the water more turbid. These conditions can adversely impact fish, including the banded killifish. The contractor will be required to follow best soil erosion and sediment control practices to minimize the loss of soil from the construction site.

2. MINIMIZATION AND MITIGATION MEASURES

2.1 Plans to Minimize Area, Estimated Number of Individuals That Will Be Taken, and Amount of Habitat Affected

The temporary causeways will only be constructed to access the two piers where the steel beams need repair. A causeway will not be needed to traverse across the entire width of the Des Plaines River. The width of the rock causeways will be as narrow as possible to support construction equipment. The total amount of temporary disturbance of river habitat (size of causeways) is 13,261 square feet.

To keep as much river habitat open as possible, only one causeway will be constructed and operated at a time. Once work is done on one side of the bridge, the temporary causeway will be removed and the other causeway could then be constructed.

In addition, temporary culverts will be placed through the causeways to allow the flow of the river to pass. This will minimize temporary ponding and obstructions of flow, keeping the flow conditions as close to existing conditions as possible.

An estimated two banded killifish could be impacted as a result of the construction. Although the fish are not likely to be present near the work zone due to noise and vibrations and are likely to swim away from the rock placement work, fish could be killed during the placement of rock for the temporary causeways. Since banded killifish reportedly travel in small schools and sample surveys typically had two or fewer banded killifish, two banded killifish are estimated to be impacted. A loss of two fish would not jeopardize the Des Plaines River population.

2.2 Plans for Management of the Area Affected That Will Enable the Continued Use of the Species

The river habitat will be restored to pre-construction conditions. There will be no long-term loss of available river habitat as a result of the proposed work. The only work in the river during construction will be the placement of the temporary rock causeways. Once work is completed, the rock causeways will be removed and the river bottom and any rock islands restored to pre-construction conditions. Fortunately, the existing channel bottom and island consist mostly of rock so the removal of the temporary rock causeways can be easily assimilated into the existing rock substrate materials.

Additional habitat improvement actions are described in Section 2.3.

2.3 Description of All Measures to be Implemented to Minimize or Mitigate the Effects on the Species

Several actions are proposed to minimize or mitigate adverse effects to the banded killifish. First, only one temporary causeway will be in place and operational at a time. This leaves habitat available on the opposite side of the river and maximizes available habitat during construction.

Culverts will allow the river to continue to flow through the causeways, minimizing flow disruptions. This will minimize flow impacts to habitat upstream and downstream of the temporary culverts as a current will be maintained closer to the shore, rather than being blocked and having to flow around the causeway.

The culverts will also minimize the formation of sediment deposits over rock substrate which can occur if the current is blocked. Sediment deposition can be harmful to fish. Also, before causeway removal, any accumulated sediment will be excavated and hauled off-site. This will minimize sediment disturbance and sediment pollution as the causeway is removed.

When the work is completed, the river habitat will be restored to pre-construction conditions. This includes the re-establishment of a rock channel bottom at the same elevations. No long-term loss of habitat is anticipated.

One habitat improvement action will be incorporated into the project to help mitigate any adverse effects. Along the shoreline within the project area, emergent plants, such as water willow, will be planted. Aquatic plants are an important habitat feature during spawning and they provide another resource for macroinvertebrates, which are the main source of food of the banded killifish. The value of work associated with these plantings is estimated to be approximately \$ 11,700.

Additionally, the Illinois Natural History Survey is seeking a \$5,000 grant for research related to the Banded Killifish, and funding of this grant will be incorporated into the project. The purpose of the proposed research is to investigate the genetic composition of expanding populations of Banded Killifish (*Fundulus diaphanus*) in Illinois with an emphasis on determining whether one or both subspecies are present, whether hybridization between subspecies are occurring, and providing management recommendations for the preservation of the species in Illinois. A copy of the research proposal has been included with this conservation plan as Exhibit C.

2.4 Plans for Monitoring the Effects of Measures Implemented

Due to Asian Carp concerns, the Lockport stretch of the Des Plaines River will continue to be a focus area for fish sampling by the IDNR, USFWS, and other agencies. The results of the sampling will be used to monitor the banded killifish population and to examine if there are any deleterious project effects. Although the exact locations of the sampling efforts can vary due to river conditions and available equipment and personnel, sampling locations are typically located immediately upstream

and downstream of the project site. The City will coordinate with these agencies to submit the results to IDNR's Endangered Species Program.

As part of its construction management duties, the City will monitor the establishment of planted vegetation along the shore. The contractor will have performance guarantees to successfully establish the emergent plants and the City will periodically review the survivability and coverage of the plants to meet contract requirements.

2.5 Adaptive Management Practices

The following adaptive management practices to deal with changed or unforeseen circumstances that may affect the banded killifish are proposed:

1. If the post-construction monitoring results are drastically reduced from the current population estimates, the City will work with the IDNR and IDOT to identify the causes, evaluate adaptive mitigation measures, and implement mitigation measures. With the increasing improvement of water quality of the Des Plaines River and the minimal construction impacts, the City does not anticipate a population loss at the project site.
2. If unforeseen construction situations occur, the City will consult with the IDNR and IDOT to make sure any corrective actions do not adversely impact the banded killifish. Potential adverse situations include flooding, equipment spills or leaks, and fuel spills.
 - a. Construction crews will monitor weather forecasts for heavy rain and flooding. If flooding is expected, all equipment will be moved out of the floodplain and construction work will be buttoned up to withstand higher flows. The temporary causeways are expected to withstand any higher flows due to the limited height, rock size, and culverts.
 - b. The release of fuels and other construction pollutants could adversely impact fish. To protect the waterways, all equipment fueling will take place at a designated location out of the floodplain and wetlands. This location will have spill response materials to clean up any accidentally spilled fuels or equipment leaks.
3. Construction workers will be informed of sensitive resources in the area, including threatened and endangered species, regulations protecting the species, where the species might be found, equipment avoidance areas, how to report sightings or incidents that may involve take, and the importance of avoiding take of the species. If any threatened or endangered species are encountered, actions will be taken to avoid impacting these species. The IDNR will be consulted on the best methods to continue work. Finally, the status report will include a map and GPS coordinates of any listed species found within the project footprint, a description of any relocations, injuries or mortalities, and the disposition of any individuals that were injured or killed.

2.6 Funding

This project will be funded jointly using federal funds (administered through IDOT) and City funds. IDOT and the City have both included this project in their FY18 capital improvement plans. There are line items specifically for this project in the FY18 budgets.

3. ALTERNATIVE ACTIONS

In addition to the proposed action, the City examined alternative actions to rehabilitate the IL 7 Frontage Road Bridge or improve habitat for the banded killifish.

3.1 Work from Bridge

For similar bridge rehabilitation projects, it is conceivable that equipment could be placed on the bridge deck and workers and materials lowered from the bridge deck. A tarp or similar device would capture any flakes and debris that falls from the bridge during construction. A temporary causeway would not need to be constructed under this alternative, therefore, there would be no work in the river. Potential impacts to the banded killifish would be limited to impacts caused by noise vibrations.

This alternative is not feasible since the bridge deck cannot safely support construction equipment. The bridge is in such poor shape that it is closed to all traffic.

3.2 Work from Barge

Construction equipment could be placed on barges and transported to the project site. However, the Des Plaines River is not suitable for barge traffic. First, the river is not maintained for navigation. Barge navigation is dedicated on the adjacent Chicago Sanitary and Ship Canal. Second, the water depths are too shallow to float a barge. Water depths at the causeway sites are mostly less than 2 feet deep so a barge would not be able to reach the work sites.

3.3 Habitat Projects

In addition to augmenting aquatic vegetation beds (see Section 2.3), three supplemental projects to increase habitat for the banded killifish were considered but dismissed. Since the presence of islands within the Des Plaines River channel help provide a variety of habitats, the City considered the establishment of additional island habitats. During removal of the temporary causeways, some rocks could be left behind and re-shaped to form channel islands for fish habitat. This is not a feasible option due to regulatory restrictions. Permit and easement restrictions from both MWRD and IDNR do not allow for the placement of any new fill or improvements within the river channel.

There is a small drainage ditch that flows into the Des Plaines River from the west side just north of the IL 7 Frontage Road Bridge. At the bridge site, most of the river channel flows rapidly. The banded killifish is known to inhabit quiet backwater areas (Section 1.2). To increase the diversity of habitat, the mouth of the unnamed tributary to the Des Plaines River could be enlarged to provide additional backwater-type habitat. This alternative was dismissed based on ecological reasons. First, the water quality is poor since the stream drains an urban area. The banded killifish are not likely to be attracted to poor water quality habitat. Second, the grading would impact a riverine wetland.

With the avoidance and minimization of wetland impacts a design criteria and the poor water quality, this alternative was not pursued.

Rock riffles could be constructed from the temporary causeway rock. This alternative was dismissed since the channel bottom is dominated by rock substrate so additional rock riffles are not needed and are duplicitous.

3.4 Fish Stocking

Fish stocking was considered but dismissed since there is not a fish hatchery that commercially produces the local genotype of the banded killifish.

3.5 No Action

Under the no action alternative, the bridge would not be rehabilitated. The bridge would remain closed, directly affecting businesses along the river and ship canal, including a proposed new transfer terminal, and the Lockport Lock and Dam. Traffic would have to follow a long detour.

The IL 7 Frontage Road used to be the original IL 7 route. As part of the construction of the new IL 7 Bridge, the IL 7 Frontage Road Bridge is to be kept open to serve the businesses and government institutions along the river and ship canal.

4. IMPACT ON LIKELIHOOD OF SURVIVAL

The proposed action will not reduce the likelihood of the survival of banded killifish in the wild in Illinois, the biotic community to which the banded killifish belongs, or the habitat essential for the species existence in Illinois.

4.1 Distribution and Habitat

As described in Section 1.2, the banded killifish is found in clear glacial lakes and as well as quiet backwaters and sluggish reaches of medium to large streams with low gradients streams and rivers. It has been found in the Mississippi, Rock, Illinois, Chicago, Kankakee, Fox, and Des Plaines River drainages (MWH and J. Meldrim, 2016).

The banded killifish consistently occurs in sampling events at the Lockport pool of the Des Plaines River. The number of banded killifish collected from each survey ranged between 0 and 161 with most survey events between 0 and 2 fish. Numerous sampling events collected more than 10 fish.

The river habitat at the fish survey areas is similar to the project site, namely an island complex habitat with rock/cobble substrate. Due to the high gradient, the velocity of the flow is relatively rapid. At the project site, the fish likely prefers the downstream side of islands as well as slower flow areas along the river banks.

4.2 Proposed Work

The potential loss of individual banded killifish as a result of the proposed work is estimated at two individuals. Based on the nearby sampling results, the population of banded killifish at Lockport could range between 20 to more than 200 individuals. A potential loss of two individuals would not likely result in adverse impacts to the banded killifish population in the lower Des Plaines River, which is expected to have a population greater than 200 individuals.

With a wide range of habitat across northern Illinois, any incidental take at the project site will not impact the statewide population of the banded killifish. The proposed project work will be kept to a minimum footprint, so fish populations outside of this area will not be effected.

The banded killifish have been found at all sampling areas on the Des Plaines River between the electric dispersal barrier at Romeoville and its confluence with the Kankakee River. With populations upstream and downstream, recruitment of banded killifish to the project site to occupy suitable habitat is likely.

The loss of habitat due to causeway construction would be temporary. During causeway removal, the river bed and any rock islands would be restored to pre-construction conditions (similar elevations and rock substrate). Therefore, river habitat conditions would be similar before and after construction.

The proposed work would also include the proposed plantings of emergent plants along the shoreline, such as water willow. This would add habitat structure for use by the fish.

5. REFERENCES

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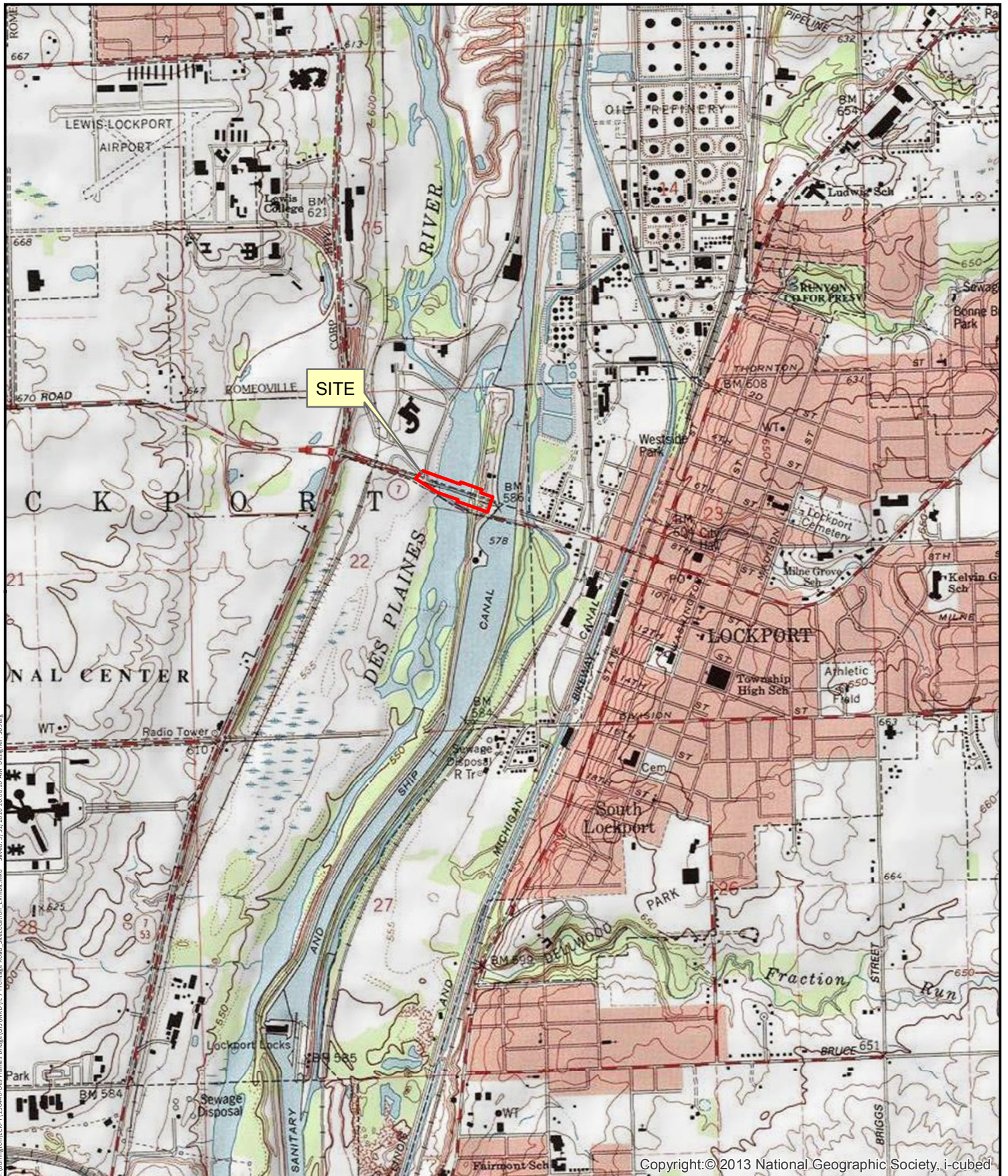
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SITE LOCATION MAP

IL 7 Frontage Road Bridge

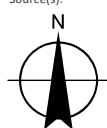
EXHIBIT A

City of Lockport, Illinois



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**CITY OF LOCKPORT
 IL 7 (W 9TH STREET)
 FRONTAGE ROAD BRIDGE**

**EXHIBIT B
 TEMPORARY CAUSEWAYS**

SCALE: 1"=40' STA. 13+50 TO STA. 22+50

F.A.U. RTE.	SECTION	COUNTY	TOTAL SHEETS	SHEET NO.
351	15-00083-00-BR	WILL	1	1
CONTRACT NO.				
FED. ROAD DIST. NO. 1 ILLINOIS FED. AID PROJECT				

Executive Summary

Application Date:

23 January 2018

Project Title:

Genetic analysis of the rapid expansion of Banded Killifish (Fundulus diaphanus) in Illinois.

Organization:

Illinois Natural History Survey

1816 S. Oak St.

Champaign, IL 61820

Grant Initiator:

Josh Sherwood (Associate Ichthyologist – Illinois Natural History Survey)

Dr. Philip Willink (Senior Research Biologist – Shedd Aquarium)

Project Purpose:

The purpose of this project is to investigate the genetic composition of expanding populations of Banded Killifish (*Fundulus diaphanus*) in Illinois with an emphasis on determining whether one or both subspecies are present, whether hybridization between subspecies are occurring, and providing management recommendations for the preservation of the species in Illinois.

Contact Information:

Josh Sherwood

Associate Ichthyologist

Illinois Natural History Survey

1816 S. Oak St.

Champaign, IL 61820

jsherwo2@illinois.edu

Funds Requested:

\$5,000

Narrative

Genetic analysis of the rapid expansion of Banded Killifish (Fundulus diaphanus) in Illinois.

Project Goals:

Recent investigations into the rapid expansion of Banded Killifish in Illinois has brought to light significant conservation implications (Willink et al., 2018). Previous data suggests that Illinois should be inhabited by the western subspecies of Banded Killifish (*F. d. menona*) (April and Turgeon, 2006), though morphological data from recently encountered populations reveals these populations may be comprised of the eastern subspecies (*F. d. diaphanus*) and/or subspecies hybrids (Willink et al., 2018). Until now, the Illinois Endangered Species Protection Board has not had to recognize subspecies. It is possible the recent expansion of Banded Killifish populations throughout Illinois may lead to the removal of its threatened status, regardless of the subspecies responsible for the expansion. The goal of this project is to provide the resource managers of Illinois a complete understanding of the expansion of Banded Killifish in Illinois by genetically determining whether these populations are comprised of the native *F. d. menona* or the non-native *F. d. diaphanus* and whether hybrids of the two are present in the state.

Objective 1:

The first objective of this study is to compare the molecular characteristics of Banded Killifish populations in Illinois to those from Minnesota (Western Banded Killifish, *F. d. menona*) and Maryland (Eastern Banded Killifish, *F. d. diaphanus*). Analyses of these data will determine whether individual populations of Banded Killifish in Illinois are comprised of the eastern or western subspecies, or potentially both. Because the western subspecies is thought to be native to Illinois, identifying populations (if any) of the eastern subspecies within the state will allow managers and decision makers to focus conservation efforts appropriately.

Objective 2:

The second objective of this study is to confirm whether or not hybridization between subspecies is occurring in Illinois, as suggested by Willink et al. (2018). By analyzing both nuclear and mitochondrial genetic markers, we will be able to identify if and where hybridization might be occurring. If present, identifying hybrid areas will further aid managers in the conservation of the appropriate subspecies of Banded Killifish in Illinois.

Needs Assessment

The Banded Killifish *Fundulus diaphanus* is a relatively wide spread cyprinodontid species. Its range stretches along the Atlantic coast of North Carolina to Nova Scotia and New Brunswick, the Great Lakes and St. Lawrence basins, and upper Midwestern states (Page and Burr, 2011). Two subspecies of *F. diaphanus* are recognized and recent research suggests limited geographic overlap between the ranges of the two subspecies (April and Turgeon, 2006). *Fundulus. d. diaphanus* (Eastern Banded Killifish) occupies the Atlantic coast and inland to Lakes Ontario and Erie, whereas *F. d. menona* (Western Banded Killifish) ranges from Lake Erie, west to Lakes Huron and Michigan, and the upper Midwest. Range-wide populations of Banded Killifish are considered secure, as well as nationally in both the United States and Canada (NatureServe, 2017). Despite the overall status being secure, populations of Banded Killifish in Wisconsin and North Dakota are considered Vulnerable, West Virginia and Iowa are Imperiled, and Manitoba, Newfoundland, South Dakota, South Carolina, and Illinois are Critically Imperiled (NatureServe, 2017).

Historical Illinois populations of the Western Banded Killifish (*F. d. menona*) were predominantly located in northeastern Illinois (Figure 1). Primarily thought to inhabit clear, well vegetated lakes (Smith, 1979), there have also been records taken from the Des Plaines River (Meek, 1910), Fox River (INHS Fish Collection #85810), Calumet River (INHS Fish Collection #84579), as well as Lake Michigan (Jordan, 1878). As lakes and wetlands were filled and multiple stressors deteriorated water bodies, the range of Banded Killifish in Illinois shrunk, until it was only found in a small number of glacial lakes (Smith, 1979). The Banded Killifish was listed as threatened in Illinois in 1989 due to restricted habitats or low populations in the state, and is still classified as such today (Illinois Endangered Species Protection Board, 2012).

Since 2000, the Banded Killifish has become more prevalent and wide-spread in Illinois (Figure 1). Historical glacial lake populations have remained relatively stable over time. Samples from Lake Michigan beach surveys first collected Banded Killifish in 2001 and it has become more wide-spread and abundant throughout the Illinois portion of the lake since. Moving inland, Banded Killifish have become widespread in the Chicago Area Waterway System (CAWS) and upper Illinois River since 2006. Finally, the first report of Banded Killifish in the Mississippi River occurred in 2009 in Davenport, Iowa, and since have spread downstream as far as West Alton, Missouri (Tiemann et al., 2015; Willink et al., 2018).

Prior to 2009, there were no known records of the Banded Killifish from the Illinois portion of the Mississippi River, nor are there known nearby populations from which this population could have originated. The appearance of this population could be explained by 1) downstream migration from Mississippi River populations in Wisconsin and Minnesota or from the Iowa and Des Moines Rivers; 2) migration from populations in the Illinois River through the defunct Hennepin Canal; and/or 3) human release. Genetic analysis of the Mississippi River population will aid in determining whether this population is due to natural migrations, or from human error, and will allow managers to act accordingly.

While the sudden and drastic increase in a threatened species is often considered good news, recent morphological analyses of the “new” populations of Banded Killifish in Illinois suggest that recent increases may not wholly be the Western Banded Killifish (Willink et al., 2018). Lateral scale rows of Banded Killifish collected in the Calumet Region appear to be intermediate between the accepted counts of *F. d. diaphanus* and *F. d. menona*, suggesting that hybridization may be occurring in Illinois waters and the potential for populations of Eastern Banded Killifish in the state. April and Turgeon (2006) noted natural hybridization of the subspecies in Lake Erie, as it is where the two subspecies potentially co-occur, yet all Banded Killifish in Illinois were hypothesized to be of the Western subspecies. Genetic analyses of the expanding Banded Killifish populations in Illinois are needed to determine whether recent expansions are due to increases in the native subspecies or an “invasion” of the non-native subspecies. These analyses are a crucial step in conserving the Western Banded Killifish in Illinois and will provide managers further data in their attempts to maintain this threatened species.

The Illinois Endangered Species Protection Board does not protect recognized subspecies, only species level classifications. Recent apparent range expansions of Banded Killifish in Illinois may result in the suggested removal of this species from the list of Endangered and Threatened Species in Illinois, possibly without considering which subspecies has expanded in the state, nor which subspecies is native. Conservation of the appropriate subspecies of Banded Killifish in Illinois will support the range-wide diversity of the species. Regardless of which subspecies is found to be the cause of the recent Banded Killifish expansion in Illinois, a genetic analysis of the current population will aid in the conservation of the species in the state. Populations of species with once limited ranges are susceptible

to reduced genetic diversity from founder effects or population bottlenecks (Meffe, 1986). Reduced genetic diversity can hinder a species' ability to adapt to changes in the environment or lead to the fixation of detrimental alleles (Meffe, 1986), both of which could have negative effects on Banded Killifish populations in Illinois.

Methods

Sample Collection

Banded Killifish will be collected from areas representing seven populations – southern Lake Michigan in Illinois and Indiana, the CAWS, northern Illinois and southern Wisconsin, the Mississippi River and lower Rock River, and the St. Joseph River of northeast Indiana and southern Michigan. If possible, individuals representing known Western (from Minnesota) and Eastern (Maryland) subspecies will be gathered and analyzed. In the absence of individuals from the 'pure' populations, genetic sequences from April and Turgeon (2006) will be accessed through GenBank. A minimum of fifteen individuals from each region will be gathered using applicable sampling methodologies for genetic analyses. Complete individuals will be euthanized and preserved in 95% EtOH. Whole individuals will be used instead of tissue samples to allow for morphological analyses (e.g. scale counts, fin ray counts, pigmentation, etc.) if evidence of genetic hybrids are discovered. Specimens will be deposited in the Illinois Natural History Survey Fish Collection, Champaign, after completion of the study.

Genetic Analyses

Genomic material will be extracted from muscle tissues via common extraction kits and protocols. Target locus for our analysis will mirror those described by April and Turgeon (2006) which consists of the mitochondrial D-loop and the nuclear X-src intron. Primer design for each locus can be found in Lee et al. (1995) (mtDNA D-loop) and April and Turgeon (2006) (nDNA X-src intron). Extracted genomic materials will be amplified via polymerase chain reaction (PCR), verified via gel electrophoresis, cleaned using standard ExoSap-IT protocols, and sequenced. Sequencing of amplified materials will be performed by Eurofins Genomics.

Assessment of genetic variation will be performed using population assignment analyses in STRUCTURE and/or Structurama2 software (or similar). These analyses will determine the number of distinct genetic groups (K) of Banded Killifish in Illinois, whereas a $K = 1$ signifies a single, panmictic population across the state and a $K > 1$ suggests multiple genetically distinct populations in the state. These analyses across both nDNA and mtDNA genes can identify evidence of hybrids among subspecies across populations within the state. A similar analysis was performed on a multi-species complex of turtles in the southern United States (Spinks et al., 2013).

Evaluation

Data from this project will be used to evaluate the current status of Banded Killifish in Illinois. Analysis of molecular data will determine the number of distinct genetic populations of Banded Killifish currently inhabiting the waters of Illinois, whether the historical subspecies is responsible for recent populations increases, as well as whether hybridization between subspecies is occurring. The potential discovery of Eastern Banded Killifish and/or subspecies hybrids in Illinois will expose the need for significant management considerations or a reevaluation of subspecies' ranges.

The results will be used to define management units that can be used to develop conservation and management plans. The Endangered Species Protection Board of Illinois will use the data to determine if the species should continue to be listed as Threatened. Although the emphasis is on Illinois, Banded Killifish distributions extend into all neighboring states, indicating that any management implications in Illinois will have a bearing on the conservation of the species in Indiana, Wisconsin, Iowa, Missouri, and beyond. The project has already garnered intense interest among ichthyologists from Indiana, Ohio, Missouri, and Wisconsin, and discussions of collaboration have begun.

Budget

*Materials**

Cost/extraction	Individuals	Genes/individual	Replicates	# of extractions	Extraction \$
\$12.04	105	2	50	260	\$3,131

Sequencing

Cost/sequence	Individuals	Genes/individual	Replicates	# of Sequences	Sequencing \$
\$3.00	105	2	50	260	\$780

Travel

Mileage	Per Diem	Lodging	Travel \$
\$361.00	\$288	\$440	\$1089

TOTAL COST \$5,000

*Materials and supplies include lab consumables (pipette tips, Eppendorf tubes, Parafilm, etc.), Extraction Kits, Primers, PCR kits, Clean-up kits, and gel electrophoresis materials.

Schedule

January – February 2018: Gather specimen already preserved by collaborators.

February 2018 – March 2018: Perform molecular extractions on collected specimen.

March – July 2018: Collect remaining specimen needed.

July – October 2018: Perform molecular extractions on last specimen.

November – December 2018: Send molecular products to be sequenced.

January – March 2019: Analyze sequence data.

April – June 2019: Prepare manuscript for peer-review publication.

Literature Cited

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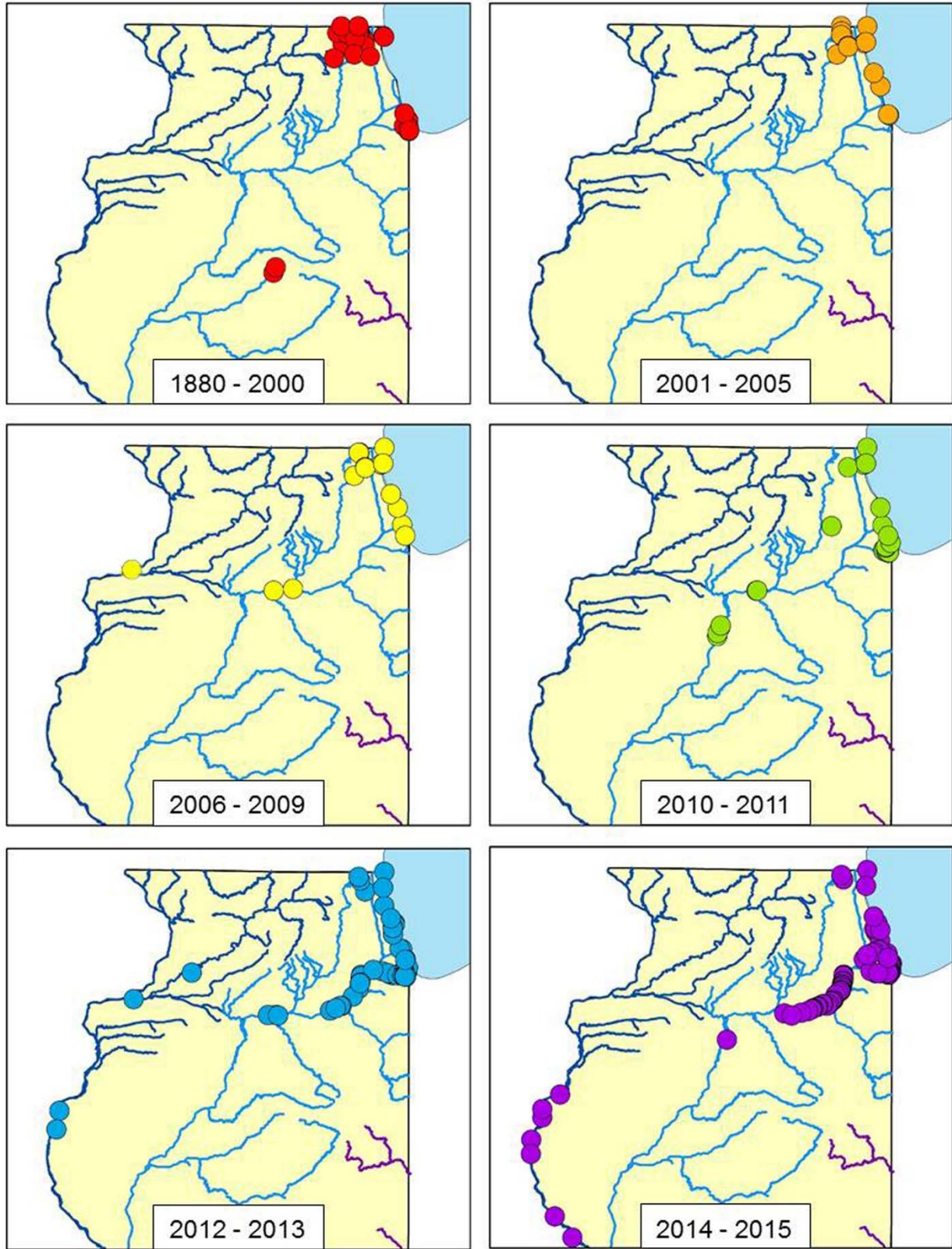


Figure 1. Changes in the range of *Fundulus diaphanus*, during select time periods, in Illinois from 1880 – 2015.



South side of bridge looking west across Des Plaines River



View of the Des Plaines River from the IL 7 Frontage Road Bridge looking east. Islands adjacent to the north of the bridge are also shown.



Looking north at Des Plaines River, islands, and shorelines



Looking south at Des Plaines River, islands, shorelines, and piers of adjacent IL 7 bridge



The westernmost pier (pier 1) is shown along with the vegetated western shoreline (facing South)



Southern face of the bridge and southwest shoreline in the distance. (facing West, photo taken in winter)



Northwest quadrant of the bridge, shoreline, and adjacent vegetation shown



One example of severe corrosion in lower webs of beams and diaphragms (Pier 2, Beam 4 Shown)

Implementation Agreement
Illinois Route 7 Frontage Road over Des Plaines River Bridge Rehabilitation
Lockport, Illinois

- A) The names and signatures of all participants in the execution of the conservation plan;

This Conservation Plan will be implemented by the City of Lockport and their duly authorized representative has signed below committing to the execution of this Conservation Plan as a part of the project.

- B) The obligations and responsibilities of each of the identified participants with schedules and deadlines for completion of activities included in the conservation plan and a schedule for preparation of progress reports to be provided to the IDNR;

The City of Lockport is solely responsible for completing this project through its designated consultants and contractors.

Construction start date: Winter 2018/2019

Construction completion date: Late Summer/Fall 2019

IDNR will be notified of the time/location of the preconstruction meeting, the start of construction, and the completion of construction. Progress reports will be provided quarterly during construction.

- C) Certification that each participant in the execution of the conservation plan has the legal authority to carry out their respective obligations and responsibilities under the conservation plan;

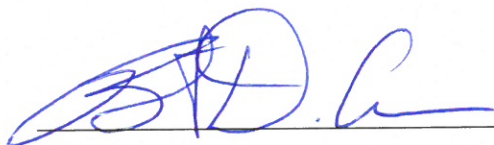
The City of Lockport hereby certifies that it has the authority and funding to complete the project and to implement all proposed conservation measures included in the Conservation Plan for the Banded Killifish.

- D) Assurance of compliance with all other federal, State and local regulations pertinent to the proposed action and to execution of the conservation plan;

The City of Lockport is in charge of construction and will assure that all applicable state, federal, and local laws will be adhered to during the completion of the project.

- E) Copies of any final federal authorizations for a taking already issued to the applicant, if any.

No federal permits for Take have been issued.



DATE: 2-26-18

Brent D. Cann, PE, MBA – Director of Public Works