Project Title: Abundance and distribution of early life stages of invasive carp in the Ohio River: 2025 Technical Report

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Participating Agencies: Kentucky Department of Fish and Wildlife Resources (KDFWR), West Virginia Division of Natural Resources (WVDNR), United States Geological Survey (USGS), United States Fish and Wildlife Service (USFWS), West Virginia University (WVU), Eastern Illinois University (EIU), Southern Illinois University (SIU), The Ohio State University (OSU).

Location of Activities: Ohio River Basin

Statement of Need:

Acquiring a full understanding of the early life history information is imperative for evaluating the population status (i.e., extent of invasion). Identifying the specific locations that differentiate both the extent of spawning and recruitment is crucial information for implementation of management or control efforts (e.g. targeted removal efforts, informing barrier or deterrent placement, etc.). In order to identify these locations, quantifying abundance and distribution of invasive carp early life stages is needed. For the purposes of this plan, the term 'invasive carp' is primarily referring to Silver Carp and Bighead Carp (*Hypophthalmichthys* species), also known as bigheaded carp, however, some work specifically on Black Carp is occurring in the lower Ohio River basin.

In order to limit the negative impacts of invasive carp populations and their further spread, efforts have increased to understand the distribution and abundance of invasive carp in the waters they currently inhabit. Previous sampling efforts on the Ohio River have documented adult invasive carp presence as far upstream as Robert C. Byrd Dam (R.C. Byrd) near Gallipolis Ferry, West Virginia. Densities of adult invasive carp are highest downstream of McAlpine Lock and Dam (Louisville, KY) and substantially decline farther upstream. In 2021, YOY were captured in Cannelton Pool (RM 691.5) and in 2022, they were captured even further upstream at RM 508.6 in Markland Pool. However, since 2016 the majority of Ohio River YOY have been consistently captured in J.T. Myers Pool.

Suspected reproduction of non-indigenous bigheaded carp, through the morphometric identification of invasive carp-type larvae, was documented in Meldahl Pool in 2016 by EA Engineering. In addition, genetically confirmed bigheaded carp eggs and larvae were collected as far upstream as Markland Pool in 2021. Previous efforts have been successful in collecting invasive carp eggs, embryos, and larvae in the Ohio River. However, defined spawning locations and the spatial extent of spawning in the Ohio River remains a knowledge gap. Multiple years of data collection covering a broader spatial extent under a variety of environmental conditions will be necessary to fully understand invasive carp early life history among pools.

To support both the national control plan (Conover et al. 2007) and the Ohio River Fish Management Team (ORFMT) Basin Framework objectives (ORFMT 2014), this project was initiated in 2016 to improve our ability to detect both early stages of invasion and spawning populations of invasive carp (Strategy 2.8) and also monitor upstream range expansion and changes in distribution and abundance (Strategy 2.3). The results of this project will help managers make informed decisions during future planning efforts regarding resource allocation for invasive carp deterrent and control strategies.

2025 Project Objectives:

1) Identify locations and the upstream extent of invasive carp spawning in the Ohio River Basin.
2) Determine the extent, biological characteristics, and environmental requirements of invasive carp recruitment locations in the Ohio River basin.
3) Evaluate drain structure operational changes to limit invasive carp recruitment in Hovey Lake.
4) Determine genetic structure and relatedness of invasive carps in the Wabash and Ohio River Basins to identify sources of propagule pressure and inform contract harvest at large-scales.
5) Refine identification of natal locations for invasive carp collected from the Ohio River Basin using otolith strontium isotope analysis, focusing on areas upstream of the establishment front.
6) Identify tributaries to the Ohio River most thermally and hydrographically suitable for invasive carp reproduction.
7) Quantify the current distribution and relative abundance of invasive carp in tributaries to the Ohio River nearest the invasion front.

Project Highlights:

- Unlike the previous two years, above average river conditions throughout May and June allowed for favorable invasive carp spawning conditions.
- A total of 123 ichthyoplankton tows were conducted in the mainstem Ohio River from J.T. Myers Pool to R.C. Byrd Pool. An additional 94 ichthyoplankton tows were conducted at tributaries or power plants along the Ohio River.
- The majority of suspect (morphometrically identified) invasive carp eggs and larvae were captured in and downstream of Cannelton Pool, with some individuals collected as far upstream as the Kentucky River in McAlpine Pool. Many suspicious larvae from RC Byrd, Greenup, and Markland Pools are still pending final genetic results.
- Targeted Young-of-Year (YOY) sampling effort included 175 surface trawl tows, 110 dozer trawl transects, and 10 electrofishing transects. YOY invasive carp were collected in J.T. Myers (N = 749), Cannelton (N = 114), Markland (N = 13), and Meldahl (N = 1) pools. The Meldahl Pool YOY Bighead Carp was the furthest upstream a YOY invasive carp has been documented.
- Targeted YOY Invasive Carp sampling occurred at four sites along the lower Ohio River. YOY Invasive Carp were found at two of the four sites and YOY Black Carp found out one of the sites. YOY Black Carp were found at two sites along the lower Mississippi River
- Reproduction of invasive carps in the Wabash River was relatively high in 2025, with the second highest larval density since 2016, and the highest egg density since 2016. Invasive carp ichthyoplankton was collected at all tributary and mainstem sampling sites, and was greatest in the Wabash River downstream of the White River confluence.

- Water strontium isotope ratio ($^{87}Sr/^{86}Sr$) data indicate that the Ohio River can be distinguished from several of its tributaries using this chemical marker, including some tributaries that are indistinguishable from the Ohio River using only Sr:Ca and Ba:Ca. Several tributaries in the Ohio River basin can also be distinguished from one another using $^{87}Sr/^{86}Sr$. Therefore, including $^{87}Sr/^{86}Sr$ analysis of otoliths in addition to analysis of Sr:Ca and Ba:Ca will provide additional resolution for inferring environmental history (including identification of natal environments) of invasive carps.
- Preliminary models using field collected temperature data and estimated water velocities suggest Silver Carp eggs are at risk of hatching in all of Ohio's major tributaries (Little Miami, Great Miami, Scioto, Hocking, and Muskingum Rivers) to the Ohio River. Risk was the lowest in the Little Miami River, but only under rare flow/temperature conditions (cold and dry years).
- No invasive carp of any life stage were observed in any of the four sampling trips conducted between May and October 2025 in any of the three locations on each of the Little Miami, Great Miami, Scioto, Hocking, and Muskingum Rivers. Although invasive carp have been periodically observed in lower reaches of these rivers through other sampling efforts, invasive carp are not common or widespread in Ohio tributaries at this time.

Methods:

For analysis purposes and for the remainder of this report, the phrase "bigheaded carps" will be referring to Silver and Bighead carps (*Hypophthalmichthys* spp.) only. YOY will be defined as fish less than 150 mm, and juvenile will define fish between 150 to 400 mm (likely 1 to 2 years old) which have undeveloped gonads and are not capable of spawning. Adult invasive carp are defined as fish greater than 400 mm with mature, identifiable gonads. Additionally, the term "suspect *Hypophthalmichthys*" is referring to an egg, advanced egg, or larvae with morphometric characteristics aligning with bigheaded carps, while the terms "suspicious egg/larvae" refers to specimens that do not have 100% of the morphometric characteristics of bigheaded carps but still warrant genetic confirmation.

Ichthyoplankton sampling:

To evaluate the extent of invasive carp spawning activity in the Ohio River above Markland L&D, West Virginia University and WVDNR conducted ichthyoplankton tows at sampling sites within the R.C. Byrd (N = 3), Greenup (N = 1) Meldahl (N = 2), and Markland (N = 2) pools. Each sampling site was visited three to four times from May 28 to June 25, 2025. During each visit, three to four tows were conducted: three within the Ohio River proper, and one additional tow within the tributary or at the intake structure if the site was a previous EA Engineering larval sampling site. Larger tributary sites (Kanawha River and Raccoon Creek) had multiple tows conducted within the tributary during each sampling event.

INDNR and KDFWR conducted ichthyoplankton sampling within tributaries within J.T. Myers (N=2), Newburgh (N = 2), Cannelton (N = 3), McAlpine (N = 2), and Markland (N = 3) pools once or twice each from May 8 to May 30, 2025, during ideal spawning conditions (water temperatures between 15.5 to 26.7°C (60 to 80°F) with moderate to high water 2-3 days after peak flow event). Additionally, the mainstem Ohio River was sampled in conjunction with tributary sites within each pool during the same timeframe. Three tows were conducted at each sampling site. INDNR also completed sampling within the East Fork, West

Fork, and mainstem White River to determine relative reproductive contribution within the White River system.

For all tows, a conical ichthyoplankton net (0.5 m, 500 µm mesh) was use. Depending on the setup, some boats deployed the net from the bow of the boat and motored in reverse. Other boats deployed the net via a push boom setup on the front of the boat and idled forward to sample. The net was pushed/pulled upstream for three minutes. Water volume sampled was recorded using a General Oceanics Flowmeter fitted to the ichthyoplankton net; depth (m) and water temperature (°C) were recorded using a boat-mounted depth sounder. All contents in the ichthyoplankton net were rinsed into a 500 µm sieve and preserved using 95% non-denatured ethanol (at an estimated ratio of nine parts ethanol to one-part sample volume) for physical identification in the lab. Suspect *Hypophthalmichthys* eggs and larvae were morphometrically identified (process outlined below) and a subsample were sent to the genetics lab at WVU for genetic confirmation.

Larval fish were initially sorted into non-invasive carp and potential invasive carp (suspicious) species using morphometric parameters provided by Auer (1982). Furthermore, early developmental characteristics outlined by Yi et al. (1998) and Chapman (2006) were utilized to physically identify suspect *Hypophthalmichthys* larvae, advanced eggs, and eggs from each sample (Figure 1). Invasive carp larvae were identified by the presence of an eye spot, and suspect *Hypophthalmichthys* were differentiated from Grass Carp (*Ctenopharyngodon idella*) and Black Carp (*Mylopharyngodon piceus*) using myomere counts. *Hypophthalmichthys* larvae have 38 to 39 myomeres, whereas Grass Carp larvae range from 43 to 45 myomeres and Black Carp have 40 and 41 myomeres. Suspect *Hypophthalmichthys* eggs were identified based on general size and presence of a large perivitelline membrane (5 to 6 mm in diameter). Suspect *Hypophthalmichthys* 'advanced eggs' were defined as the beginning of a yolk-sack larvae still contained within the perivitelline membrane. In some cases, eggs and/or larvae were labeled as "suspicious" if they exhibited some (but perhaps not all) morphometric characteristics of invasive carp. Out of caution due to the collection location of the suspicious specimens, they were pulled and sent to the genetics lab for genetic confirmation of species.

FluEgg Modeling:
In 2025, the U.S. Geological Survey Ohio–Kentucky–Indiana Water Science Center, in cooperation with the INDNR, began development of a FluEgg model of the Ohio River between McAlpine Lock and Dam and Smithland Lock and Dam. The effort focuses on simulating the transport, dispersion, and development of silver carp eggs spawned within the study reach to estimate where environmental conditions support successful maturation to the gas bladder inflation stage. Work started with customizing the National Weather Service community HEC-RAS hydraulic model for the purposes of this study. Following the customization, model performance was evaluated by comparing simulated water-surface elevations to observed elevations for selected historical events to ensure that the hydraulic conditions driving egg transport were reasonably represented.

Wabash River ichthyoplankton:
Eastern Illinois University (EIU) sampled ichthyoplankton in the Wabash River mainstem and four of its tributaries to monitor invasive carp (*Hypophthalmichthys* spp.) reproduction. EIU used a bow-mounted ichthyoplankton push net (Wildco), 0.5 meters in diameter, 2.5 meters in length, and 500 µm mesh. The tributaries sampled include the Vermilion, White, Embarras, Little Wabash Rivers. At each tributary nine

push net samples were collected: three within the tributary, three in the Wabash River above the tributary, and three in the Wabash River below the tributary. Each triplicate of samples consisted of a left bank, middle, and right bank sample to cover multiple areas across the channel. Each push net sample lasted five minutes and the volume of water filtered was estimated by a General Oceanics flow meter to achieve a target sample volume of 50 cubic meters. The contents of the net were emptied into a sample container and preserved with 95% non-denatured ethanol for identification in the lab. Invasive carp eggs and larvae were identified using meristic and morphometric features. Samples containing extremely high densities of invasive carp larvae and/or eggs were randomly split down to a proportional subsample of ½ - 1/16th total sample volume to allow feasible sorting amounts. A subsample of invasive carp eggs and larvae were submitted for genetic analysis. A handheld YSI multiparameter meter was used to record temperature (°C). River discharge data (m^3/s) from gauges nearest to sampling locations were obtained from United States Geological Survey.

Targeted YOY Sampling:
Targeted YOY invasive carp sampling using surface and dozer trawls and took place in tributaries and embayments of the Ohio River from J.T. Myers Pool to Racine Pool. Due to YOY Silver and Bighead Carp being found in Markland Pool in 2022, crews put significantly more effort into sampling upstream tributaries annually since then.

The surface trawl measured 3.7 m wide, 0.6 m tall, and 5.5 m deep with 31.8 mm bar (number 12) netting. An additional layer of 4.8 mm mesh (35-pound delta) bag was attached externally to improve capture of small fishes. Additional foam floats were added to the top line of the trawl to provide extra buoyancy. Otter boards were 30.5 cm tall, 61.0 cm long, and each had a 12.7 cm diameter, 27.9 cm long "buoy style" PVC float attached to the top of the board allowing them to float. The trawl was deployed off the bow of the boat and attached with 24.4 m ropes. The boat was motored at 1.6 to 3.2 km per hour in reverse for five minutes before retrieving the net. In some locations it was not possible to complete five minutes of trawling, in which case sample time was documented. At the biologist's discretion, additional trawls were conducted at sites where either coverage was limited, or juvenile invasive carp were suspected. In addition to surface trawls, pulsed-DC electrofishing (64 hz, 37% duty cycle) was conducted in various tributaries of Markland Pool. All invasive carp were identified to genus, measured to total length, and weighed.

An electrified dozer trawl was used in the upper Ohio River for additional targeted YOY sampling. The dozer trawl net was attached a frame and boom system and deployed off the bow of the boat just below the water's surface. The frame measured 7ft wide and 3ft deep with an 18ft long net. The net mesh was 38mm stretched, HDPE body and 4mm heavy duty raschel netting. A transect was completed travelling upstream using electricity for 5min at approximately 3.2km/h. Sample time was documented if a full 5-minute run could not be completed. Electrofishing settings were based on conductivity to achieve a target power goal for targeting invasive carp based on unpublished data and Young et al. (2023) for targeting invasive carp with electrofishing. Processing of samples followed similar methodology as the surface trawl.

Black Carp YOY Sampling:
KDFWR conducted targeted sampling for YOY Black Carp in the lower Ohio River from the confluence with the Mississippi River to above Smithland lock and dam and in the lower Mississippi River from the confluence of the Ohio River to the lower border of the state. Sampling locations were chosen based on the hydrologic similarity to the location where YOY Black Carp were collected previously in Kentucky.

Sampling effort did not exceed 30 days. When YOY Black Carps were collected, length was recorded, and the specimens were preserved for additional analysis as needed. Most sites were sampled using a backpack electrofisher (Smith-Root LR-24) for variable durations depending on amount of habitat available to sample. Sampling with 20' x 5' and 15' x 5' (1/8" mesh) seines were used where environmental conditions allowed.

Hovey Lake recruitment:

Hovey Lake is a known nursery area within the lower Ohio River, with YOY finding their way into the lake nearly every year. Excluding flood events, all water passing into and out of Hovey Lake must funnel through three culvert pipes at a control structure across Bayou Drain. Blocker boards can be installed within the culvert pipes during certain times of year to control the amount of water entering or leaving the lake. This control structure is operated by the Hovey Lake Fish and Wildlife property staff to manage the lake level primarily for waterfowl hunting opportunities. From May 29, to July 14, 2025, multiple gears were used weekly within Bayou Drain on both the river side and lake side of the water control structure to evaluate the timing of YOY passage from the Ohio River into Hovey Lake.

Sampling for larval invasive carp was conducted using a conical ichthyoplankton net (0.5m, 500 µm mesh) deployed from the front of the boat. The boat was motored in reverse, pulling the ichthyoplankton net for three minutes (approximately 180 meters). This was repeated three times per side of the control structure. The water volume sampled was recorded using a General Oceanics Flowmeter fitted to the ichthyoplankton net; water temperature (^{o}C) was recorded using a boat-mounted depth sounder. All contents in the ichthyoplankton net were rinsed into a 500 µm sieve and preserved using 95% non-denatured ethanol (at an estimated ratio of nine parts ethanol to one-part sample volume) for physical identification in the lab. Suspect and suspicious *Hypophthalmichthys* larvae were sorted from all other native fish larvae and were counted and measured.

A modified, rigid-frame surface trawl (herein called a beam trawl) was developed specifically for sampling in Bayou Drain with a small jon boat. Due to accessibility issues, a small jon boat was the only option for sampling the lake side of Bayou Drain. The standard 3.7 m wide surface trawl with otter boards that is used for other Ohio River YOY sampling was too large and produced too much drag to be able to effectively pull it with a small jon boat. The beam trawl consisted of a wooden frame measuring1.83 m wide and 0.61 m tall, with a 3.66 m long bag of 4.8 mm mesh (35-pound delta). Additional weight was added to the bottom of the frame to properly orient the trawl such that it floated upright in the water and just below the surface when towed. The trawl was deployed off the bow of the boat and attached with 20 m tow ropes. The boat was motored at 1.6 to 2.6 km per hour in reverse for the duration of the trawl before retrieving the net. A target goal of 14 minutes of trawling covering approximately 730 m was conducted on each side of the control structure each day. Individual tows lasted 3.5 to 5 minutes depending on obstacles in the water. After each tow, fish were sorted and YOY invasive carp were counted and measured.

In April 2023, USGS installed a stream gage within Bayou Drain that monitors stream stage and velocity to compute streamflow in Bayou Drain. Adjustments and calibrations were made throughout 2024, but the water flow near the control structure is challenging and gives variable results. Because of this, the velocity meter on this station was removed in 2025, only allowing for gage height to be determined.

Wabash River Genetics:

EIU collaborated with an external lab (Dr. Devon Keeney at LeMoyne College) to process genetics samples for species identification and to investigate the genetic structure of invasive carp early life stages in the Wabash River Basin. A subsample of larvae and eggs collected in 2025 were sent to Dr. Keeney's lab for species identification.

Strontium isotope analysis

Water samples for strontium isotope ratio ($^{87}Sr/^{86}Sr$) analysis were collected from 8 Ohio River navigation pools (Olmsted, Smithland, J.T. Myers, Newburgh, Cannelton, McAlpine, Markland, and R.C. Byrd) and 29 tributaries during summer and fall 2025. Five Wabash River tributaries (White, East Fork White, West Fork White, Patoka, and Little Wabash rivers) were also sampled. Sampling locations included major tributaries where invasive carp reproduction has been documented or may occur based on suitable habitat, as well as smaller tributaries of the Ohio River where juvenile bigheaded carps have been captured in recent surveys. Water samples were filtered, strontium extracted, and analyzed for $^{87}Sr/^{86}Sr$ using a thermal ionization mass spectrometer at the University of Wisconsin-Madison. Water $^{87}Sr/^{86}Sr$ data were compiled with existing water Sr:Ca and Ba:Ca data for the Ohio River and tributaries to characterize multivariate water chemistry 'signatures' for locations across the Ohio River basin and determine which locations can be distinguished from one another using these three natural chemical markers in combination.

Sectioned lapilli otoliths from silver carp collected from the Ohio River and tributaries by Indiana DNR and other agency field crews during 2021-2022 that were previously analyzed for Sr:Ca and Ba:Ca will be used in this study. Prioritization of otolith samples to be analyzed for $^{87}Sr/^{86}Sr$ will be based in part on uncertainty of early life assignments for individual fish yielded by otolith Sr:Ca and Ba:Ca analysis (prioritizing fish for which otolith microchemistry could not identify a specific early life environment) and inclusion of samples from fish collected from each of the five Ohio River navigation pools (J.T. Myers, Newburgh, Cannelton, McAlpine, and Markland) for which we have otolith samples with microchemistry data. Otolith samples will be analyzed for $^{87}Sr/^{86}Sr$ using multi-collector, laser ablation ICPMS. The laser will be used to ablate a spot in the otolith core (targeting otolith growth from the fish's early life history) adjacent to the previous laser ablation conducted to analyze samples for Sr:Ca and Ba:Ca. Otolith core $^{87}Sr/^{86}Sr$ data will be used in conjunction with previously obtained otolith core Sr:Ca and Ba:Ca data to refine early life environment assignments for individual fish. Otolith analysis for this objective has been delayed but is expected to be completed in 2026.

Modeling risk assessment of invasive carp reproduction in Ohio tributaries

Ohio State University (OSU) deployed 18 temperature loggers longitudinally across the Little Miami, Great Miami, Scioto, Hocking, and Muskingum Rivers in April 2025. These loggers are continuously maintained, and data is downloaded every 2-3 months. OSU also modeled predicted water velocity profiles and other hydrologic measures from the Hydrologic Engineering Center's River Analysis System (HEC-RAS) for the five target tributaries. HEC-RAS models were developed for the continuous river lengths starting at locations identified as potential spawning habitat for Silver Carp. We assumed the most downstream dams on the Great Miami, Scioto, and Muskingum Rivers would be barriers to Silver Carp and provide high velocity habitat for spawning, so we defined spawning areas just downstream of these dams (Figure 2). The Little Miami and Hocking Rivers are smaller and dispersal to the most downstream dam in these systems is unlikely. Therefore, the most downstream high velocity area in these systems

were used as spawning locations (Figure 2). Temperature and water velocity data for our target reaches in each river were needed as input into FluEgg models to assess reproductive potential.
Bigheaded carp have been observed spawning from March to September, therefore, we considered a wide range of environmental conditions when creating FluEgg models. We considered twenty-five scenarios per tributary where discharge was either set at the fifth, twenty-fifth, fiftieth, seventy-fifth, or ninety-fifth percentiles and temperature was set at eighteen, twenty-one, twenty-four, twenty-seven, and thirty degrees Celsius. We only used discharge during the potential spawning period (March to September) to calculate percentiles, and minimum and maximum temperatures were set based on the minimum required temperature for bigheaded carp spawning (18C) and the approximate maximum daily average temperature observed (30C). FluEgg takes the user inputs and applies internal processes to account for the effects of changing water density and egg development on egg buoyancy. Egg buoyancy, along with 3-dimensional estimates of water velocity, are used to predict the vertical, lateral, and longitudinal position of eggs in the water column at a given time. We considered any egg that was in the lower fifth percentile of the water column at the time of hatching to be settled out (i.e., non-viable). Therefore, the proportion of eggs (n = 1000) in the upper ninety-fifth percentile was used as a metric of hatching risk.

Ohio river tributary sampling

OSU sampled the Great Miami, Little Miami, and Scioto Rivers via boat-electrofishing at three fixed sites in each river four times from May to September (n = 24). All sites were within ~100 km upstream of the Ohio River confluence and are spaced relatively equidistant. River access is limited and natural barriers to boat traffic (e.g., rapids) exist; therefore, sites were established upstream of access points starting at the first barrier. The sampling methodology is a modified version of the EPA's NRSA protocol for boatable rivers. OSU sampled 7,000 seconds across eight equal-length littoral subreaches starting on a randomized bank and alternating bank every two subreaches. During low water events, a portion of some of the most upstream subreaches could not be accessed and effort was scaled to match the length of river sampled. During high water events, high velocities in certain subreaches made it impossible to reach the effort goal, so additional effort was applied in slower reaches to again scale effort to length of river sampled. Fishes were processed after every two subreaches, identified to species, and measured to the nearest millimeter. Smaller individuals were preserved in 10% formalin and identified in the lab.

Results and Discussion:

Ichthyoplankton sampling:

A combined total of 66 ichthyoplankton tows were conducted across nine sites within the mainstem Ohio River in R.C. Byrd (N = 12), Greenup (N = 9), Meldahl (N = 18), and Markland (N = 27) pools (Table 1; Figure 3). Further downstream, a total of 57 ichthyoplankton tows were conducted at 10 different sites within the mainstem Ohio River in the J.T. Myers (N = 12), Newburgh (N = 9), Cannelton (N = 24), and McAlpine (N = 12) pools (Table 1). Suspicious and/or suspect invasive carp eggs or larvae were identified from nearly all mainstem sampling sites and were sent to the WVU genetics lab to be genetically confirmed. Preliminary genetic results suggest spawning in upper pools, however final results are still pending as of April 15, 2026. Unless otherwise noted, all egg, advanced egg, and larvae counts listed herein are referring to morphometrically identified *Hypophthalmichthys* species. Significantly more larvae were captured throughout the Ohio River in 2025 than in the past several years. Of note, Cannelton Pool accounted for the highest number of eggs (N = 1,682), Newburgh Pool accounted for the greatest number of advanced eggs (N = 1,869) and J.T. Myers accounted for the majority of larvae collected (N = 1,135).

An additional 94 ichthyoplankton tows were conducted in 19 tributaries of the Ohio River (Table 1; Figure 3). Four tributaries in Markland Pool and further upstream (Hogan Creek, Little Miami River, Scioto River, Kanawha River) had suspicious larvae that were sent off for genetic confirmation. In J.T. Myers Pool, 65 eggs and 40 larvae were captured in the Green River, and approximately 44,621 post gas-bladder larvae were collected in Bayou Drain of Hovey Lake. One suspicious larvae was captured in both the Anderson River of Newburgh Pool and the Blue River of Cannelton Pool, and one suspect egg was collected in the Salt River of Cannelton Pool. In McAlpine Pool, 53 eggs were captured in the Kentucky River. Lastly, the White River (a Wabash River tributary) is known to produce invasive carp eggs and larvae, but more precise spawning reaches in the White River system are unknown. Three ichthyoplankton tows were conducted on each the mainstem White River, West Fork White River, and East Fork White River. The West Fork produced substantially more suspect eggs (95/1,000 m^3), advanced eggs(322/1,000 m^3), and larvae (1,147/1,000 m^3) than the East Fork (14 eggs/1,000 m^3, 2 advanced eggs/1,000 m^3, 2 larvae/1,000 m^3. The mainstem White River had approximately half the egg and larvae densities of the West Fork, indicating that the West Fork White River is likely the major contributor of invasive carp reproduction occurring in the White River basin.

Unlike 2023 and 2024, Ohio River reproduction appeared to be substantially higher in 2025. River conditions were the likely driver behind the substantial increase in spawning. River stage remained well above average during the spring spawning window in 2025, and experienced numerous peak flow events throughout that time, unlike the previous two years (Figure 4). The vast majority of the spawning continues to be taking place downstream of McAlpine L&D, however, more upstream locations detected spawning than in previous years. The majority of the spawning in the Ohio River appears to be occurring in the mainstem river. Limited reproduction is occurring in tributaries, however, the Green River and Kentucky River appear to have some amount spawning occurring in them (excluding Smithland and Olmstead pool tributaries). Genetic results are still pending final analysis. Preliminary results suggested Silver Carp spawning much further upstream than previously detected, so select samples are being reevaluated for accuracy. A final report summarizing genetic results will be shared with the partnership as soon as possible.

FluEgg Modeling:
Following hydraulic model evaluation, FluEgg simulations were conducted to estimate egg dispersion from known and suspected spawning events at McAlpine Lock and Dam during spring flow conditions in 2020, 2021, 2022, 2023, and 2024. Provisional simulation results were reviewed with INDNR personnel and used to identify scenarios of for further analysis. These simulations are currently in progress. A USGS report summarizing methods and results is planned for release later this year or early next.

Wabash River ichthyoplankton:
EIU collected a total of 204 ichthyoplankton samples from April 30th to July 24th, 2025, across 11 sampling dates. Samples were taken from each tributary and corresponding mainstem Wabash River sites up to seven times throughout the season, depending on access. The Vermilion River was sampled on three occasions and was inaccessible after June 11th, the corresponding mainstem site was sampled one

additional occasion in June. Following June Sampling events, most tributaries became inaccessible for the remainder of the season but select tributaries were still sampled into late July.

In total, 46,555 larvae and 16,840 eggs were visually identified as invasive carp (Table 2). A sub-sample of all eggs and larvae collected throughout the season were submitted to LeMoyne college and only partial results are available at this time (see *Wabash River Genetics* subsection). Thus, the totals above are likely a slight overestimation of invasive carp eggs and larvae produced in the Wabash River Basin in 2025.

All sites had visually identified invasive carp eggs found in the samples, with the greatest number collected in the Wabash River mainstem (n = 9,698) near the White River confluence, and from the Embarrass River tributary site (n = 4,141; Table 2, Figure). We observed distinct maximum densities of invasive carp eggs in several locations on the first day of sampling, suggesting a synchronized spawning event among multiple tributaries and the mainstem Wabash River, focused in the middle of our study reach. In the previous sampling year, genetic analysis revealed some eggs classified as probable invasive carp species due to similar size and characteristics came back as other species (e.g. freshwater drum). Thus, we used a larger size threshold to classify potential invasive carp eggs (~3mm or greater). High densities of invasive carp eggs in the White River and mainstem sites are similar to results in previous years, and further suggests this system supports a significant portion of the invasive carp reproduction in the Wabash River annually. The Little Wabash River did not produce significant numbers of invasive carp eggs in 2025 as in previous years, however the peak in discharge in this system fell just before our first sampling event so we may have missed some spawning activity in that system.

Invasive carp larvae were found in all tributaries except for the Vermilion River, with the greatest number produced in the White River (n = 2,169) and the adjacent downstream mainstem site (n = 41,982; Table 2, Figure 6). This is similar to the egg results in this system, and suggests spawning continued in this system throughout the season, as peaks in larval abundance occurred approximately six weeks after peaks in egg abundance. Overall abundance of invasive carp larvae Little Wabash River was lower than most years, but still had the second highest density of larvae in 2025. Although we may have missed some early spawning and capture of eggs early in the season, the high larval density in and around the Little Wabash River confluence suggests this continues to be a hot spot for carp reproduction as observed in prior years. The Embarrass River exhibited relatively low densities of invasive carp larvae, suggesting this tributary and confluence may be an important spawning location (e.g. high egg density), but spawning may not occur very far into the tributary as observed by the lack of larval production throughout the season. Very low numbers of invasive carp larvae were detected in the Wabash River near the Vermilion River confluence (n = 10-12), and no invasive carp larvae were found in the Vermilion River itself.

We determined a continuous sampling event to capture an entirety of a spawning event was unfeasible for various reasons, thus we focused on modelling the duration of spawning throughout a spawning season, using larval data collected from 2021-2024. We used a loess smoothed average function (non-parametric regression) to estimate the initiation of spawning (15% max), the middle of spawning (50%) max, and the peak of spawning (85%; Figure 7). From those we were able to estimate the duration of spawning as well as the peak reproduction date (Julian date), to compare among tributary and mainstem habitats. Through these analyses we determined the duration of spawning is much more narrow in tributaries (~ 10 days) when compared to mainstem habitats (~20 days; Figure 8). We also determined the peak reproduction date of invasive carp in tributaries fell later in the year (~Julian date 180, late June), compared to mainstem habitats (Figure 9). This information will be used in future analyses and will be particularly valuable in determining the best times to monitor for invasive carp early life stages, and gives further information on spawning patterns throughout a spawning season.

Our results for the 2025 sampling season showed reproduction of invasive carps in terms of egg abundance and density was higher than any other data collection year to date. In terms of invasive carp larvae, 2025 was the third greatest year in total count, and second greatest in density. Results of 2024 sampling were similar with high abundance of both invasive carp eggs and larvae collected in the drift of mainstem and tributary sites, suggesting both 2024 and 2025 were high reproductive years throughout the Wabash River Basin. The White River, Little Wabash River, and their adjacent mainstem sites were large contributors to the abundance and density of invasive carp larvae in the basin. These results are similar to previous work published by EIU that demonstrate larger tributaries with greater mean discharge throughout the spring and summer (e.g. Little Wabash and White Rivers) support significantly higher reproduction than smaller tributaries such as the Vermilion River (Schaick et al. 2020).

Additionally, we were able to identify trends in the reproductive duration and peak spawning date of invasive carp that are important for future objectives and analyses. We found a greater duration of spawning in the mainstem Wabash River compared to tributaries, suggesting favorable spawning conditions occur for a longer period in the mainstem Wabash River versus tributary habitats. This further suggests the Wabash River is a hotspot for invasive carp reproduction and potential recruitment. We found significant overlap between mainstem and tributary habitats in the peak larval production of invasive carps, with a slightly earlier peak in the mainstem Wabash River. This result is somewhat surprising given tributary habitats should warm faster than mainstem habitats, but this may be an effect of localized discharge patterns rather than temperature alone. These analyses involving spawning duration and peak spawning date will be particularly important for our future objective of describing recruitment patterns throughout the basin. With this information in hand we will be able to better predict when recruitment could be occurring and will help guide juvenile sampling efforts.

Targeted YOY Sampling:
Among project partners, targeted YOY sampling consisted of 175 surface trawl tows (11.5 hours of sampling), 110 electrofied dozer trawl transects (13.6 hours), and 10 boat electrofishing transects (4.8 hours). The majority of the effort was expended in Markland, Meldahl, Greenup, and R.C. Byrd pools (Table 3). Despite the upstream efforts, most YOY bigheaded carp were captured in J.T. Myers (N = 749) and Cannelton (N = 114) pools, with an additional 13 captured in Markland Pool and a single Bighead Carp from Locust Creek in Meldahl Pool (Table 3). The Meldahl Pool fish marks the furthest upstream documented YOY in the Ohio River basin since this project's inception (Figure 10). Of the 877 YOY invasive carp captured, 674 were Silver Carp, 132 were Bighead Carp, and the remaining 71 were unknown bigheaded carp too small to confidently confirm species.

Targeted YOY catch rates indicate that 2025 was the highest recruitment year in the past 5 years. Both J.T. Myers and Cannelton pools had well above average catch rates (Table 4). Of note, KDFWR fall fish community sampling in Cannelton Pool produced 15 additional juvenile Silver Carp ranging from 145 to 223 mm in Oil and Clover Creek of Cannelton Pool, and more were observed in Deer Creek and Little Blue. For the second time in the past four years, Markland Pool produced YOY. The single Bighead Carp YOY captured in Meldahl Pool shifts the extent of recruitment further upstream. Interestingly, Bighead carp accounted for 15% of all YOY captured in 2025 despite adult catch rates being much lower. The above average flows in May and June allowed for an extended ideal spawning window, which contributed to high recruitment in 2025.

Some additional work remains to be done to evaluate sampling effectiveness among the various targeted YOY gears. In 2024, boat speed appeared to be an important factor in determining Gizzard Shad catch (a surrogate since YOY invasive carp are rarely caught upstream), and it was recommended that surface trawling efforts use 2 mph as a target for sampling speed. However, due to data collection discrepancies in 2025 among crews, we were unable to reevaluate efforts and could not accurately determine shad catch rates (density) variation to determine if all project partners gears were sampling similarly.

Black Carp YOY Sampling:
KDFWR sampled for YOY Invasive Carp at 4 sites along the lower Ohio River, 9 sites along the Mississippi River, and no sites along the lower Tennessee and Cumberland Rivers (Table 5). YOY Black Carp were collected, in two locations along the Mississippi River and one along the lower Ohio at the confluence of the Cumberland River (Figure 11). All sites were adjacent backwaters to the mainstem of the river, less than one meter deep with sandy/muddy substrate. The location at the confluence of the Cumberland River and Ohio River, has been sampled for the past four years with YOY Invasive Carp collected each year, but YOY Black Carp have only been collected in two of the years; the 2023 Black Carp was 36 mm long, sampled on the 27th of September and the 2025 Black Carp was 33 mm long, sampled on the 26th of September. A total of 146 YOY Black Carp were collected from sites on the Mississippi River.

Efforts in 2025 revealed the presence of YOY Black Carp at three locations out of 13 sites sampled along the lower Ohio, Mississippi, Tennessee, and Cumberland River. The two capture locations on the lower Mississippi River were at mile markers 916 and 906, and the Ohio River capture location were at mile marker 920. The 2025 sites have similar habitat characteristics to historic capture locations. All sites were close to the main river channel, shallow (< 1 m), muddy and sandy backwaters. These areas appear to be serving as nursery areas at higher water levels but become isolated during lower river stages. This sampling effort has been modified to target certain backwaters where YOY Invasive Carp have been found previously. New sites are identified each year with varying environmental conditions, but many main backwaters along the Ohio and Mississippi River remain consistent across years. This occurrence is further evidence of consistent, albeit low, Black Carp reproduction in the lower Mississippi River and Ohio River drainage in Western Kentucky.

Hovey Lake recruitment:
Bayou Drain of Hovey Lake was sampled on both sides of the water control structure once a week from May 29th to July 14th, 2025, via ichthyoplankton netting and surface trawling. Ichthyoplankton netting effort consisted of 21 tows on the river side of the control structure and 28 tows on the lake side. Larvae were detected on the river side of the drain on the first day of sampling (May 29) via ichthyoplankton net. Due to the extremely large catches in some ichthyoplankton tows, numbers of larvae had to be approximated for a few samples. Approximately 46,604 post gas bladder inflation *Hypophthalmichthys* larvae were collected via ichthyoplankton nets on the river side of the drain, 96% of which were collected on May 29th. On the lake side of the control structure, approximately 26,025 larvae were collected. All but 25 of these were captured on June 23rd, which was the first day larvae were detected on the lake side. Additionally on June 23rd, water was flowing over and through the control structure into the lake (Figure 12). An ichthyoplankton

net was held stationary at the structure for 10 minutes while water flowed into the lake, which produced approximately 36,000 larvae. A subset of specimens were submitted to the WVU genetics lab for confirmation, but results are still pending.

Beam trawls efforts consisted of 47 tows, including 20 on the river side of the water control structure and 27 tows on the lake side. Shorter transects were conducted on the lake side do to downed trees blocking the way. Beam trawls first detected invasive carp on the river side of the control structure on June 4th, but did not collect any on the lake side of the structure until June 30th. Excluding June 23rd (the day high water was flowing over the structure into the lake), beam trawl catch rates increased weekly on the river side of the structure until peaking on July 7th (Table 6). Beam trawls captured YOY invasive carp between 9 and 61 mm. A prolonged spawn season produced larvae that continually found Bayou Drain over the course of the seven weeks of sampling. Even on July 14th, sampling collected YOY from 15 to 61mm long, and mean lengths were always greater on the lake side of the structure suggesting a steady new supply of younger fish coming in from the river.

The 2025 field season highlighted the fact that we only have so much control over YOY carp's ability to get into Hovey Lake. Repetitive up and down river levels, and high water that overtop the control structure make it extremely difficult to keep small invasive carp out of the lake. Unfortunately, it appears that the years that it will be most difficult to control are years when spawning output will be the highest. As previous reports have stated, a highly successful spawning event could quickly shift the current known extent of recruitment to pools farther upstream. Therefore, the spatial and temporal variation in invasive carp recruitment in the Ohio River emphasizes the need for continued long-term monitoring with this project as well as others within the basin. Efforts in this project provide valuable insight into factors promoting the reproduction and recruitment of invasive carp, and ultimately range expansion. Results support several Basin Framework and National Plan strategies and will be used by biologists to mitigate the spread of these invasive fishes.

Wabash River Genetics:
LeMoyne College continues to process our genetics samples for species identification and to investigate the genetic structure of invasive carp early life stages in the Wabash River Basin. A subsample of 2025 suspected invasive carp eggs have been analyzed and results are summarized in Table 7. The majority of suspected *Hypophthalmichthys* spp. eggs were confirmed as Silver Carp, with minor proportions of Bighead Carp, and Grass Carp. This is similar to previous data showing a mixture of these three species in high density egg samples, showing these three species continue to spawn simultaneously in the Wabash River. These proportions will be extrapolated to the full sample totals to better classify the contribution of Silver, Bighead and Grass carp egg abundance in the greater dataset. Additionally a small number of suspected invasive carp eggs came back as Silver Chub (n = 17) and Skipjack Herring eggs (n = 6). These samples were stand-alone examples of incorrectly identified samples and invasive carp eggs were not detected at all within these samples. Silver Chub eggs have a large diameter and similar morphology to invasive carp eggs, and our previous research has shown a small proportion of suspected invasive carp eggs being genetically identified as this species. Skipjack Herring eggs do not usually show up in our genetic testing, thus this is a new result we will have to look out for when visually identifying eggs in the future.

Results of our genetics study continue to show Silver Carp are the primary invasive carp species spawning in the Wabash River, although there is some simultaneous spawning of Bighead Carp and Grass Carp as well. It is important to note that just a subsample of eggs have been analyzed from the 2025 field season, so this is not a full depiction of genetic results. Faculty and students at LeMoyne college will continue to genetically identify eggs collected in 2025, and then will start to analyze larval samples. Once genetic identification is complete, we will develop the larger analysis of genetic structuring and diversity of invasive carps throughout the Wabash River Basin.

Strontium isotope analysis:
We found differences in water $^{87}Sr/^{86}Sr$ between the Ohio River and several of its tributaries, as well as among tributaries (Figure 13). Tributaries for which addition of $^{87}Sr/^{86}Sr$ will aid in distinguishing them from the Ohio River (beyond use of only Sr:Ca and Ba:Ca) include the Cumberland, Green, East Fork White, Blue, Great Miami, and Kanawha rivers and Indian Creek. There are only nine small tributaries (Little Pigeon, Oil, Little Blue, Hogan, Tanners, Deer, Sinking, Fourteen Mile, and Indian-Kentuck creeks) whose multivariate chemical 'signature' strongly overlaps with that of at least one section of the Ohio River. All major tributaries extending from the mouth of the Ohio River upstream to the Kanawha River and all major tributaries of the Wabash River are distinguishable from the Ohio River using a combination of $^{87}Sr/^{86}Sr$, Sr:Ca, and Ba:Ca. Thus, major tributary spawning locations within the current range of invasive carps in the basin can be distinguished from the Ohio River, which will enable assessment of the role of tributaries as recruitment sources for invasive carps in the basin using otolith chemistry. There are also several pairs of tributaries in the Ohio River basin that are indistinguishable from one another using Sr:Ca or Ba:Ca but differ in $^{87}Sr/^{86}Sr$. Thus, for fish identified as being of tributary origin, addition of $^{87}Sr/^{86}Sr$ analysis will enable assignment of individual fish to a particular tributary or to a small grouping of 2-5 tributaries; this resolution is not possible using only Sr:Ca and Ba:Ca. There are also differences in $^{87}Sr/^{86}Sr$ and Sr:Ca among different sections of the Ohio River that will also be useful in the context of inferring environmental history of invasive carps from otolith chemistry analyses. In general, $^{87}Sr/^{86}Sr$ is higher in upriver compared to downriver pools, whereas Sr:Ca is higher between the mouths of the Wabash and Scioto rivers compared to downstream of the mouth of the Wabash River and upstream of the Scioto River. Thus, addition of $^{87}Sr/^{86}Sr$ analysis of otoliths will substantially improve the resolution of natal environment assignments for individual invasive carp sampled from the Ohio River basin.

Modeling risk assessment of invasive carp reproduction in Ohio tributaries
Preliminary FluEgg models suggested more than 50% (and often more than 75%) of eggs spawned in the Great Miami, Scioto, Hocking, and Muskingum Rivers were predicted to be viable under all combinations of water temperature and discharge scenarios (Figures 14-17). Eggs were predicted to drift out of tributaries and into the mainstem Ohio River in 48%, 4%, 20%, and 100% of scenarios in the Great Miami, Scioto, Hocking, and Muskingum Rivers, respectively (Figures 14-17). High discharge and low water temperatures, as expected, slow down egg development and led to eggs reaching the Ohio River before hatching. The Little Miami River was the only tributary in which some scenarios led to predicted failed recruitment. In scenarios with low discharge and low water temperatures, less than 25% of eggs were predicted to hatch, with values near zero for the coldest and driest conditions (Figure 18).
Our preliminary models suggest Ohio tributaries to the Ohio River at likely to contribute to Silver Carp recruitment under the assumptions modeled so far. However, there are several next steps we plan to take to refine models for a more robust assessment. Currently, our HEC-RAS models are not fully integrated with conditions in the Ohio River mainstem. This means we had to make a simplifying assumption that

water velocities at the bottom of each reach near the confluence with the mainstem continued downstream into the Ohio River. We are currently working on patching together HEC-RAS output so we can more accurately represent confluence dynamics and water velocities in the mainstem river. We would also like to include more scenarios in our assessment, especially a wider variety of spawning locations. Some of the low-head dams or riffles assumed to be barriers to upstream movement may be passable during higher water conditions. We therefore plan to include a wider variety of spawning scenarios as we continue to refine and expand our models. As the invasion front continues to move upstream in the Ohio River, this risk assessment suggests there are several tributaries that are thermally and hydrologically suitable for Silver Carp egg development, suggesting these tributaries could be major contributors to invasive carp biomass in the Ohio River.

Ohio river tributary sampling

OSU successfully completed four sampling trips as planned on each of the three target tributaries (Great Miami, Little Miami, and Scioto Rivers). Despite our large sampling effort, we did not observe any invasive carp, including any life stage, during our sampling. This suggests invasive carp are likely not widely established and are rare in Ohio tributaries. This is good news given the predicted reproductive potential of these large tributaries for bigheaded carp and the recovering native fish communities in these rivers.

OSU characterized temporal and longitudinal variation in native fish communities while sampling for invasive carp. Across all sample events and locations, we captured a total of 78 species of fish, with Emerald Shiner, Gizzard Shad, and Bluntnose Minnow as the three most abundant species (Table 8). Statistical analyses are currently underway to quantify variation in fish community structure across the sampling season and across the sampling sites and to determine which species drive variation in community structure. This data will provide baseline information on variation in fish community structure that can serve as a reference if invasive carp should become established in Ohio tributaries and to document potential impacts on fish communities as the invasion front continues to move upstream.

Tables and Figures:

Table 1. Summary of ichthyoplankton tows collected by WVU, WVDNR, KDFWR, and INDNR. Sampling took place between May 8 and June 25, 2025.

Pool	Location	Transect Type	Tows (N)	Suspect IC Eggs (N)	Suspect IC Adv. Eggs (N)	Suspect IC Larvae (N)	Suspicious Eggs (N)	Suspicious Larvae (N)
Smithland	East Fork White River	Tributary	3	6	1	1	50	
Smithland	West Fork White River	Tributary	3	39	134	476		
Smithland	White River	Tributary	3	16	68	252		
JT Myers	Hovey Lake Drain	Tributary	6			44,621		948
JT Myers	RM 840.6 (near Hovey Drain)	Ohio River	6	73	22	198		
JT Myers	Green River	Tributary	6	65		40	113	
JT Myers	RM 784.0 (near Green River)	Ohio River	6	771	776	937		
Newburgh	Little Pigeon Creek	Tributary	3				1	
Newburgh	RM 772.8 (near Little Pigeon)	Ohio River	3	28		2		
Newburgh	Anderson River	Tributary	6			0		
Newburgh	RM 731.3 (near Anderson Rv)	Ohio River	6	223	1,896	220		
Cannelton	Deer Creek	Tributary	6					
Cannelton	RM 718.7 (near Deer Creek)	Ohio River	6	1,189	344	139		
Cannelton	Blue River	Tributary	6			0		1
Cannelton	RM 662.9 (near Blue River)	Ohio River	6	491	228	110		
Cannelton	Salt River	Tributary	6	1				
Cannelton	RM 630 (near Salt River)	Ohio River	6	1		2		2
Cannelton	RM 608.5 (New Albany ramp)	Ohio River	6	1		1		
McAlpine	Harrods Creek	Tributary	6					
McAlpine	RM 595.8 (near Harrods Creek)	Ohio River	6					
McAlpine	Kentucky River	Tributary	6	53				
McAlpine	RM 545.8 (near Kentucky River)	Ohio River	6			1		
Markland	Craigs Creek	Tributary	3					
Markland	RM 529.5 (near Craigs Creek)	Ohio River	3					39
Markland	Laughery Creek	Tributary	3					
Markland	RM 498.5 (near Laughery Creek)	Ohio River	3				1	23
Markland	Hogan Creek	Tributary	3					5
Markland	RM 496.7 (near Hogan's Creek)	Ohio River	9					5
Markland	Great Miami River	Tributary	3					
Markland	RM 490.5 (near Great Miami)	Ohio River	3				1	10
Markland	Little Miami River	Tributary	3					1
Markland	RM 463.5 (near Little Miami)	Ohio River	9					4
Meldahl	J.M. Stuart	Tributary	3					
Meldahl	RM 404.7 (J.M. Stuart)	Ohio River	9					4
Meldahl	Scioto River	Tributary	3					1
Meldahl	RM 356.4 (near Sciota River)	Ohio River	9					12
Greenup	Guyandotte River	Tributary	3					
Greenup	RM 305.2 (near Guyandotte)	Ohio River	9					17
RC Byrd	Raccoon Creek	Tributary	3					
RC Byrd	Kanawha River	Tributary	12					25
RC Byrd	RM 265.1 (Point Pleasant)	Ohio River	12					11

Table 2. Sampling location, transect location (Wabash River upstream, Wabash River Downstream, Within Tributary), number of push net samples, number of invasive carp eggs collected, mean egg CPUE (fish/m^3), number of invasive carp larvae collected, larvae CPUE (fish/m^3) collected in the Wabash River Basin in 2025. All samples are pending genetic testing and are subject to change.

Sampling Location	Transect Location	Push Net (n)	Eggs (n)	Egg CPUE	Larvae (n)	Larvae CPUE
Vermilion	Wabash Upstream	12	164	0.25	10	0.01
Vermilion	Wabash Downstream	12	288	0.48	11	0.02
Vermilion	Tributary	9	1	0.00	0	0.00
White	Wabash Upstream	21	3,920	4.57	703	0.53
White	Wabash Downstream	21	5,778	6.52	41,982	3.09
White	Tributary	9	1	0.00	0	0.00
Embarras	Wabash Upstream	18	373	0.46	24	0.03
Embarras	Wabash Downstream	18	1,127	1.77	30	0.03
Embarras	Tributary	21	4,141	4.22	326	0.13
Little Wabash	Wabash Upstream	18	174	0.19	775	0.48
Little Wabash	Wabash Downstream	18	160	0.15	473	0.51
Little Wabash	Tributary	15	113	0.13	52	0.06
Totals		**204**	**16,840**	**1.92**	**46,555**	**0.52**

Table 3. YOY sampling effort (hr) and catch (N) for various pools of the Ohio River, 2025.

Pool	Surface Trawl			Dozer Trawl			Electrofishing			Total		
	Runs (N)	Effort (hr)	YOY (N)	Runs (N)	Effort (hr)	YOY (N)	Runs (N)	Effort (hr)	YOY (N)	Runs (N)	Effort (hr)	YOY (N)
JT Myers	7	0.6	749							7	0.6	776
Newburgh										0	0.0	0
Cannelton	22	1.8	114							22	1.8	114
McAlpine	8	0.4	0							8	0.4	0
Markland	84	4.2	5	25	7.7	8	10	4.8	0	119	16.7	13
Meldahl	30	2.5	1							30	2.5	1
Greenup	24	2.0	0							24	2.0	0
RC Byrd				61	4.0	0				61	4.0	0
Racine				24	1.9	0				24	1.9	0
Grand Total	175	11.5	869	110	13.6	8.0	10	4.8	0.0	295	29.9	877

Table 4. Combined surface trawl and dozer trawl efforts (hours) and CPUE (YOY/hour) for YOY bigheaded carp in pools of the Ohio River since 2019.

Pool		Year							LTA
		2019	2020	2021	2022	2023	2024	2025	'19-'24
JT Myers	Effort	0.2	2.1	1.6	0.2	0.7	3.1	0.6	1.3
	CPUE	1490.0	293.0	714.3	228.0	0.0	0.3	1284.0	454.3
Newburgh	Effort	0.0	0.0	0.7	0.0	0.0	2.1	0.0	0.5
	CPUE	0.0	0.0	1.4	0.0	0.0	0.0	0.0	0.2
Cannelton	Effort	6.2	4.2	2.3	1.8	3.2	4.3	1.8	3.6
	CPUE	0.0	0.0	14.1	14.3	0.0	0.0	63.9	4.7
McAlpine	Effort	0.0	0.0	0.7	0.0	0.0	0.9	0.4	0.3
	CPUE	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Markland	Effort	0.0	0.0	0.0	2.1	4.7	2.3	11.9	1.5
	CPUE	0.0	0.0	0.0	6.5	0.0	0.0	1.1	1.1
Meldahl	Effort	0.0	0.0	0.0	0.0	2.4	3.1	2.5	0.9
	CPUE	0.0	0.0	0.0	0.0	0.0	0.0	0.4	0.0
Greenup	Effort	0.0	0.0	0.0	0.0	0.9	1.5	2.0	0.4
	CPUE	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
RC Byrd	Effort	0.0	0.0	0.0	0.0	1.0	9.1	4.0	1.7
	CPUE	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Racine	Effort	0.0	0.0	0.0	0.0	0.3	6.2	1.9	1.1
	CPUE	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

Table 5. Summary of YOY invasive carp and Black Carp captures in Western Kentucky.

2022	Sites Sampled	Sites with YOY Invasive Carp	Sites with YOY Black Carp
Ohio River	21	2	1
Mississippi River			
Tennesse/Cumberland Rivers	2		
Total	23	2	1
2023	**Sites Sampled**	**Sites with YOY Invasive Carp**	**Sites with YOY Black Carp**
Ohio River	24	3	1
Mississippi River	9	2	
Tennesse/Cumberland Rivers	22		
Total	55	5	1
2024	**Sites Sampled**	**Sites with YOY Invasive Carp**	**Sites with YOY Black Carp**
Ohio River	3	3	
Mississippi River	12	8	2
Tennesse/Cumberland Rivers			
Total	15	11	2
2025	**Sites Sampled**	**Sites with YOY Invasive Carp**	**Sites with YOY Black Carp**
Ohio River	4	2	1
Mississippi River	9	6	2
Tennesse/Cumberland Rivers			
Total	13	8	3

Table 6. Number and size of YOY invasive carp captured via beam (surface) trawl in Bayou Drain of Hovey Lake, 2025.

Date	Lake side of control structure				River side of control structure			
	YOY IC (N)	Length (mm)			YOY IC (N)	Length (mm)		
		Mean	Min	Max		Mean	Min	Max
5/29/2025	0				0			
6/4/2025	0				906	13.1	11	15
6/10/2025	0				4,499	17.5	14	21
6/23/2025	0				0			
6/30/2025	1,939	19.8	11	41	6,120	13.6	9	17
7/7/2025	512	35.6	26	54	14,033	23.0	18	29
7/14/2025	37	47.2	23	61	60	24.1	15	32

Table 7. Results of invasive carp egg genetics samples analyzed to date from the 2025 field season. Locations include samples taken from the mainstem Wabash River near tributaries (above and below), and within tributaries (White River, Little Wabash River, Embarras River, and Vermilion River).

Sampling location		Date Sampled	# of samples	Silver Carp	Bighead Carp	Grass Carp	Silver Chub	Skipjack Herring
White River	Below	4/30/25	23	21	1	1	0	0
(Mainstem	Above	4/30/25	30	29	1	0	0	0
Wabash River)	Above	5/12/25	3	0	0	0	0	3
	Above	5/27/25	1	0	0	0	1	0
White River	Tributary	4/30/25	3	0	0	0	1	0
Little Wabash	Below	4/30/25	3	3	0	0	0	0
(Mainstem	Above	4/30/25	3	3	0	0	0	0
Wabash River)	Below	5/12/25	2	0	0	0	2	0
	Above	5/12/25	3	0	0	0	1	2
Little Wabash	Tributary	4/30/25	3	3	0	0	0	0
Embarras	Below	4/30/25	10	9	0	1	0	0
(Mainstem	Above	4/30/25	3	3	0	0	0	0
Wabash River)	Above	5/12/25	3	0	0	0	3	0
Embarras	Tributary	4/30/25	30	28	0	2	0	0
Vermilion	Below	5/13/25	3	0	0	0	2	1
(Mainstem	Above	5/13/25	1	0	0	0	1	0
Wabash River)	Below	5/26/25	3	0	0	0	3	0
	Above	5/26/25	3	0	0	0	3	0

Table 8. Total count of all fish species encountered while sampling the Scioto, Great Miami, and Little Miami Rivers across all sites and sampling events (n=24).

Common Name	**Scioto River**	**Great Miami River**	**Little Miami River**	**Total**
Banded Darter	3	2	0	**5**
Bigeye Chub	5	0	0	**5**
Bigmouth Buffalo	2	0	0	**2**
Black Buffalo	12	0	0	**12**
Black Crappie	10	13	2	**25**
Black Redhorse	1	108	0	**109**
Blackside Darter	0	2	0	**2**
Blackstripe Topminnow	0	1	0	**1**
Blue Catfish	16	3	6	**25**

Blue Sucker	14	1	1	**16**
Bluegill	61	386	48	**495**
Bluntnose Minnow	240	1164	17	**1421**
Bowfin	0	2	0	**2**
Brook Silverside	14	31	0	**45**
Bullhead Minnow	146	139	6	**291**
Central Stoneroller	6	43	0	**49**
Channel Catfish	180	185	85	**450**
Common Carp	96	260	42	**398**
Creek Chub	1	1	0	**2**
Emerald Shiner	3202	1609	956	**5767**
Fathead Minnow	0	1	0	**1**
Flathead Catfish	76	27	12	**115**
Freshwater Drum	707	362	439	**1508**
Gizzard Shad	1131	1221	912	**3264**
Golden Redhorse	36	1148	14	**1198**
Grass Carp	1	1	2	**4**
Grass Pickerel	0	1	0	**1**
Gravel Chub	21	2	3	**26**
Green Sunfish	19	87	8	**114**
Greenside Darter	0	2	0	**2**
Highfin Carpsucker	4	9	0	**13**
Hybrid Striped Bass	39	40	16	**95**
Johnny Darter	0	1	0	**1**
Largemouth Bass	23	159	3	**185**
Longear Sunfish	130	784	49	**963**
Longnose Gar	148	56	38	**242**
Mimic Shiner	65	256	163	**484**
Mooneye	19	0	6	**25**
Mosquitofish	1	7	0	**8**
Northern Hogsucker	6	240	0	**246**
Northern Logperch	5	37	1	**43**
Orangespotted Sunfish	7	158	0	**165**
Pumpkinseed Sunfish	1	1	0	**2**
Quillback	69	107	44	**220**
Rainbow Darter	2	0	0	**2**
Redear Sunfish	14	8	0	**22**
River Carpsucker	467	504	151	**1122**
River Redhorse	8	16	3	**27**
River Shiner	3	14	7	**24**
Rock Bass	0	68	0	**68**
Rosyface Shiner	0	42	0	**42**
Sand Shiner	562	96	76	**734**

Sauger	31	27	50	**108**
Saugeye	4	2	2	**8**
Shortnose Gar	3	1	0	**4**
Silver Chub	4	12	39	**55**
Silver Redhorse	14	18	29	**61**
Silver Shiner	0	309	0	**309**
Skipjack Herring	11	0	2	**13**
Slenderhead Darter	6	1	0	**7**
Smallmouth Bass	38	322	12	**372**
Smallmouth Buffalo	721	112	243	**1076**
Smallmouth Redhorse	91	207	35	**333**
Spotfin Shiner	363	617	8	**988**
Spotted Bass	126	35	17	**178**
Spotted Sucker	0	232	1	**233**
Steelcolor Shiner	37	40	0	**77**
Streamline Chub	3	0	0	**3**
Striped Bass	0	1	0	**1**
Striped Shiner	1	62	0	**63**
Suckermouth Minnow	7	4	2	**13**
Tippecanoe Darter	2	0	0	**2**
Walleye	0	2	0	**2**
Warmouth	1	3	0	**4**
White Bass	90	63	37	**190**
White Crappie	1	1	5	**7**
White Sucker	0	18	0	**18**
Yellow Perch	0	3	0	**3**

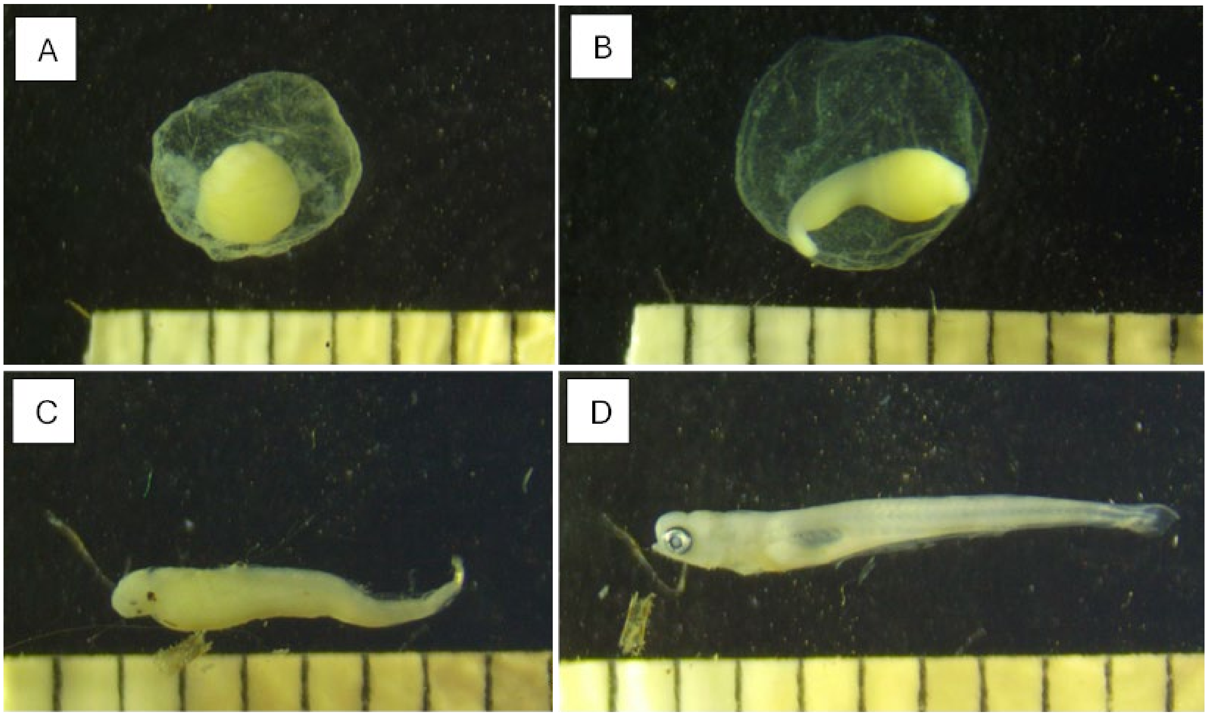

Figure 1. Developmental life stages of *Hypophthalmichthys* spp. with size comparisons. For the purposes of this report, pictures A, B, C, and D demonstrates specimens categorized as "eggs", "advanced eggs", and "yolk sac larvae" and "post gas-bladder inflation larvae", respectively.

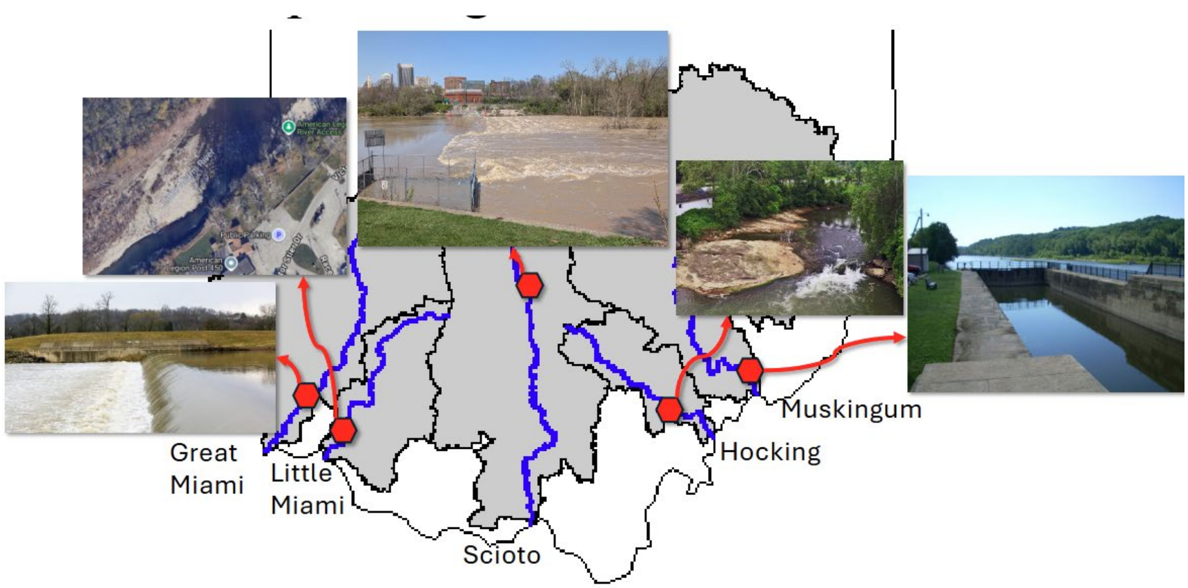

Figure 2. Hypothesized spawning locations used in Flu-Egg models to assess risk of successful Silver Carp recruitment in Ohio tributaries to the Ohio River.

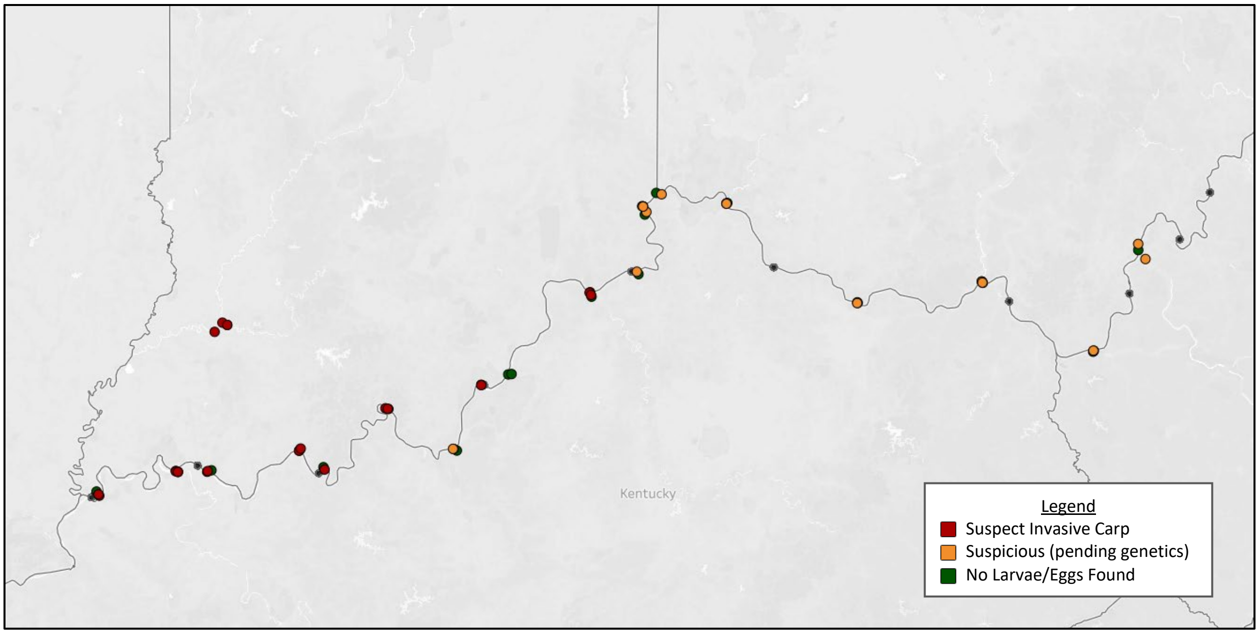

Figure 3. Map of 2025 study area of larval sampling sites. Black dots denote a lock and dam, green circles indicate larval sampling sites where no suspect or suspicious carp eggs/larvae were found, red circles indicate locations where morphometrically identified suspect *Hypophthalmichthys* eggs, embryos, or larvae were collected, and orange circles indicate where suspicious eggs or larvae were collected.

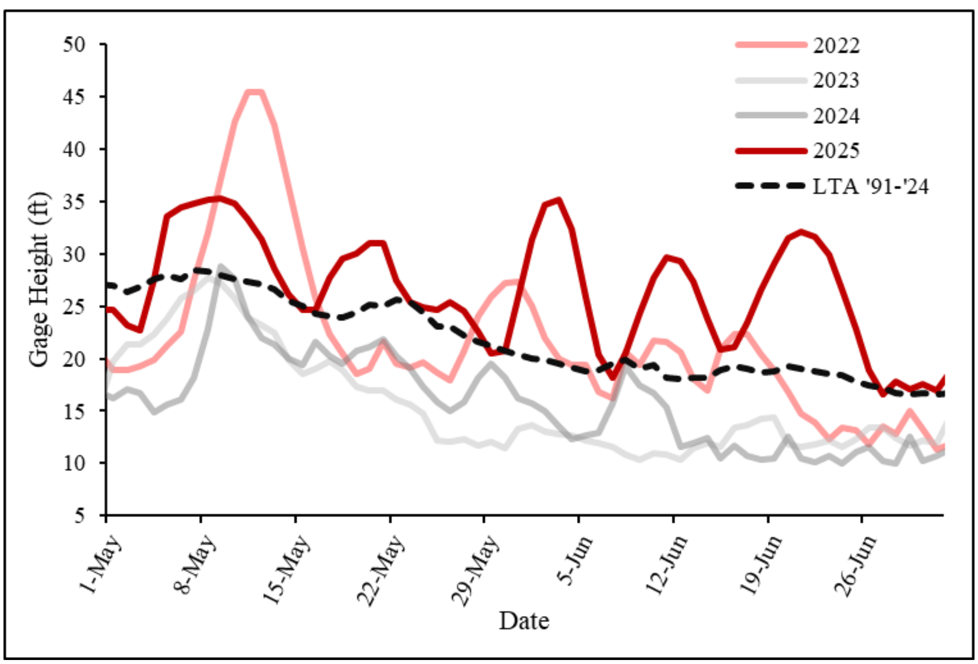

Figure 4. Annual recorded gage height (feet) for the Ohio River at Louisville, KY (USGS-03294500) from 2022 to 2025, during the primary Silver Carp spawning window from May 1 to June 30. The dark red line indicates 2025 river stage and the black dashed line indicates calculated long-term average flow from 1991 to 2024.

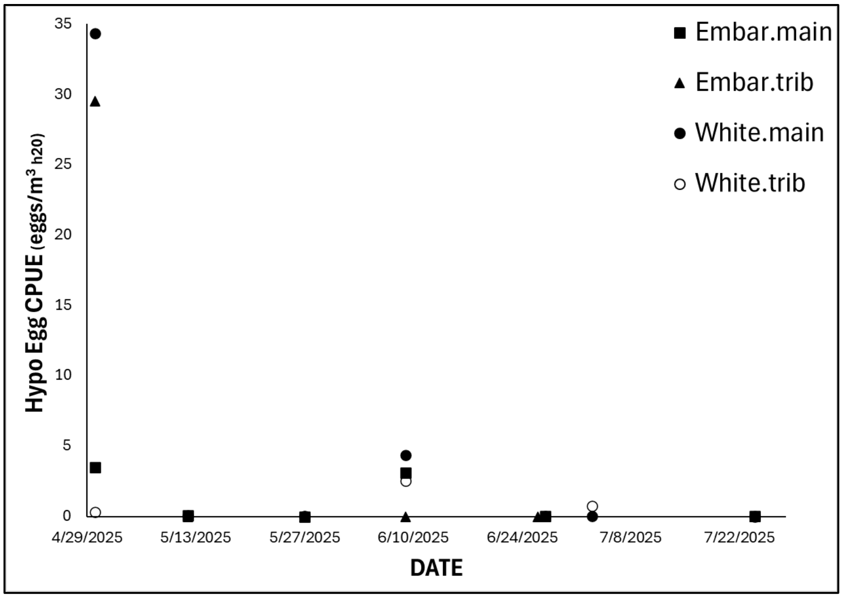

Figure 5. Catch per unit effort (eggs/m^3 H2O filtered) of invasive carp eggs collected from April 30th, 2025-July 24th, 2025 in the Embarras River (Embar.trib) and adjacent mainstem site (Embar.main), and the White River (White.trib) and adjacent mainstem site (White.main). Mainstem samples were located above and below respective tributary confluences on the Wabash River.

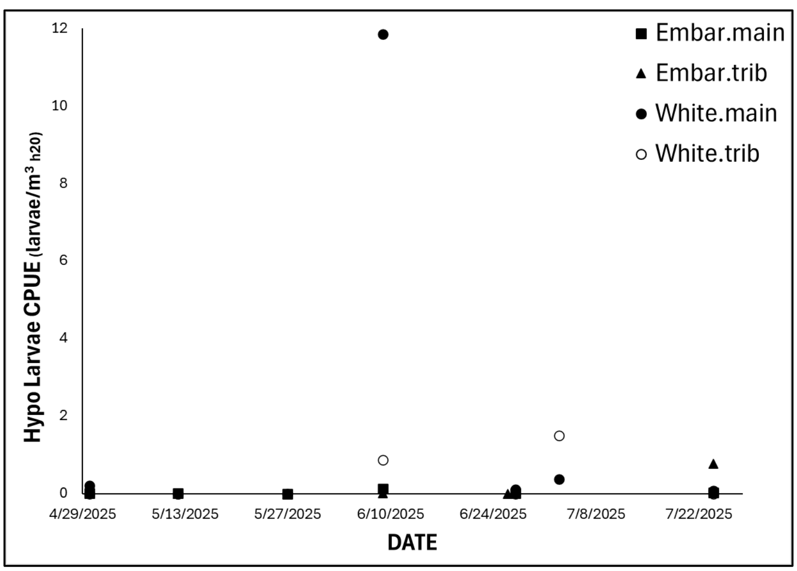

Figure 6. Catch per unit effort (larvae/m^3 H2O filtered) of invasive carp larvae collected from April 30th, 2025-July 24th, 2025 in the Embarras River (Embar.trib) and adjacent mainstem site (Embar.main), and the White River (White.trib) and adjacent mainstem site (White.main). Mainstem samples were located above and below respective tributary confluences on the Wabash River.

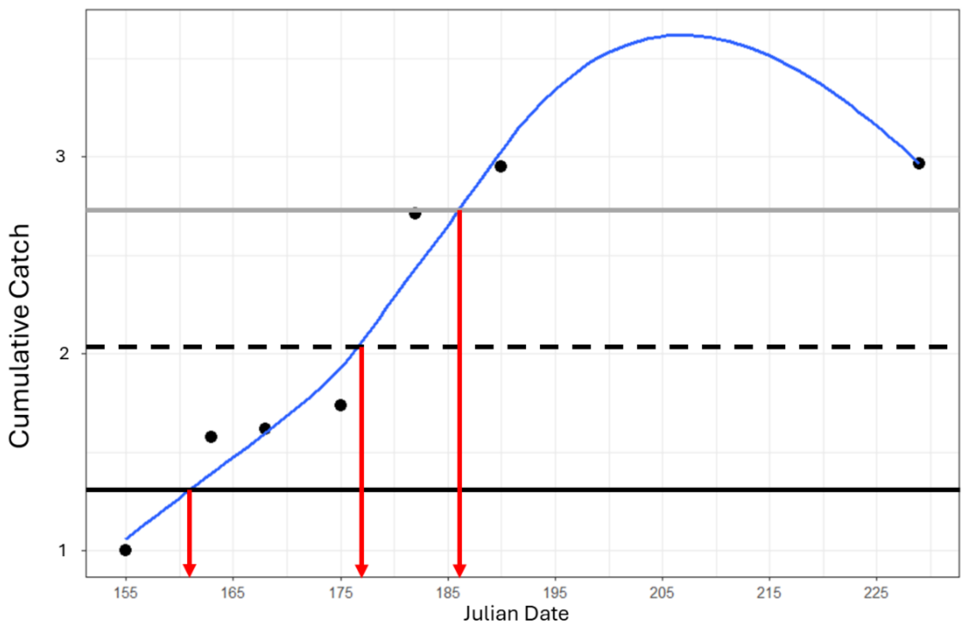

Figure 7. Cumulative larval catch curve with a loess smoothed average function. The loess smoothed average function (blue line) is a nonparametric regression used on datasets with large gaps between data points or lots of zeros. Red arrows depict 15, 50, and 85% of the cumulative larval catch. The left most arrow indicates the beginning of the spawning period (15%), middle arrow the peak (50%), and left most arrow the end of the spawning period (85%).

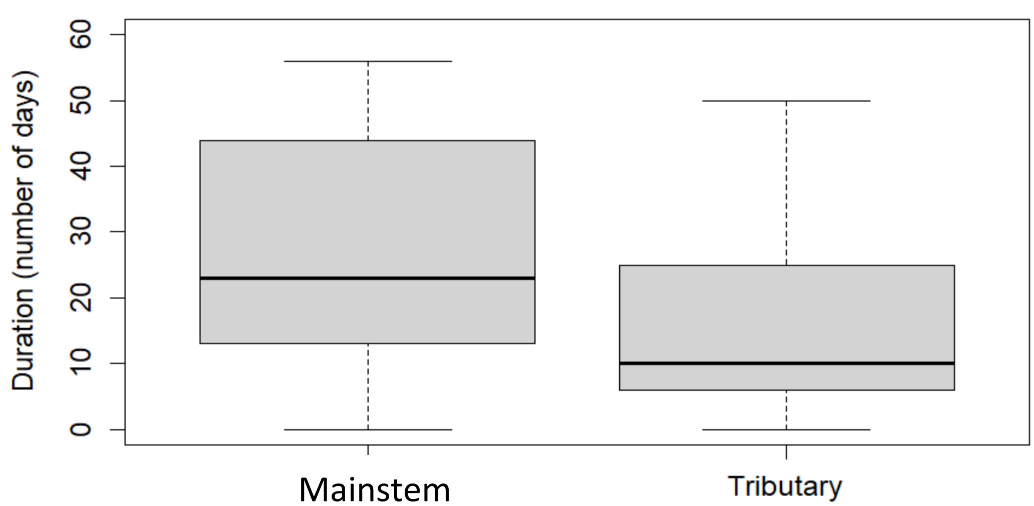

Figure 8. Box and whisker plot depicting the reproductive duration (number of days) of invasive carp reproduction (larvae) on y-axis compared among mainstem Wabash River sites and tributaries to the Wabash River on x-axis.

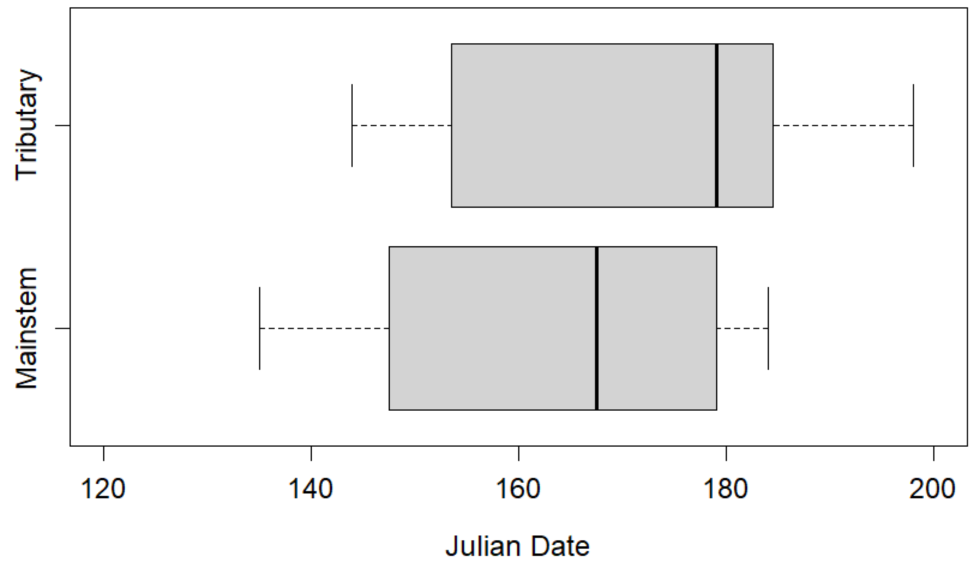

Figure 9. Box and whisker plots depicting the peak of invasive carp reproduction (larval) based on Julian Date (x-axis), among Wabash River mainstem sites, and tributaries to the Wabash River (y-axis).

Figure 10. Map of 2025 targeted YOY sampling sites. Black circles denote a lock and dam, green squares indicate sampling sites, orange squares indicate locations where YOY *Hypophthalmichthys* were collected.

Figure 11. Capture locations of YOY Invasive Carp and Black Carp along the lower Ohio and Mississippi Rivers in Western Kentucky.

Figure 12. A photo of the Hovey Lake drain water control structure on June 23, 2025, when a river rise caused water to backflow over and through the structure into Hovey Lake. The river height at JT Myers L&D (USGS-03322420, Ohio River at Uniontown Dan, KY) was at 31.1 ft.

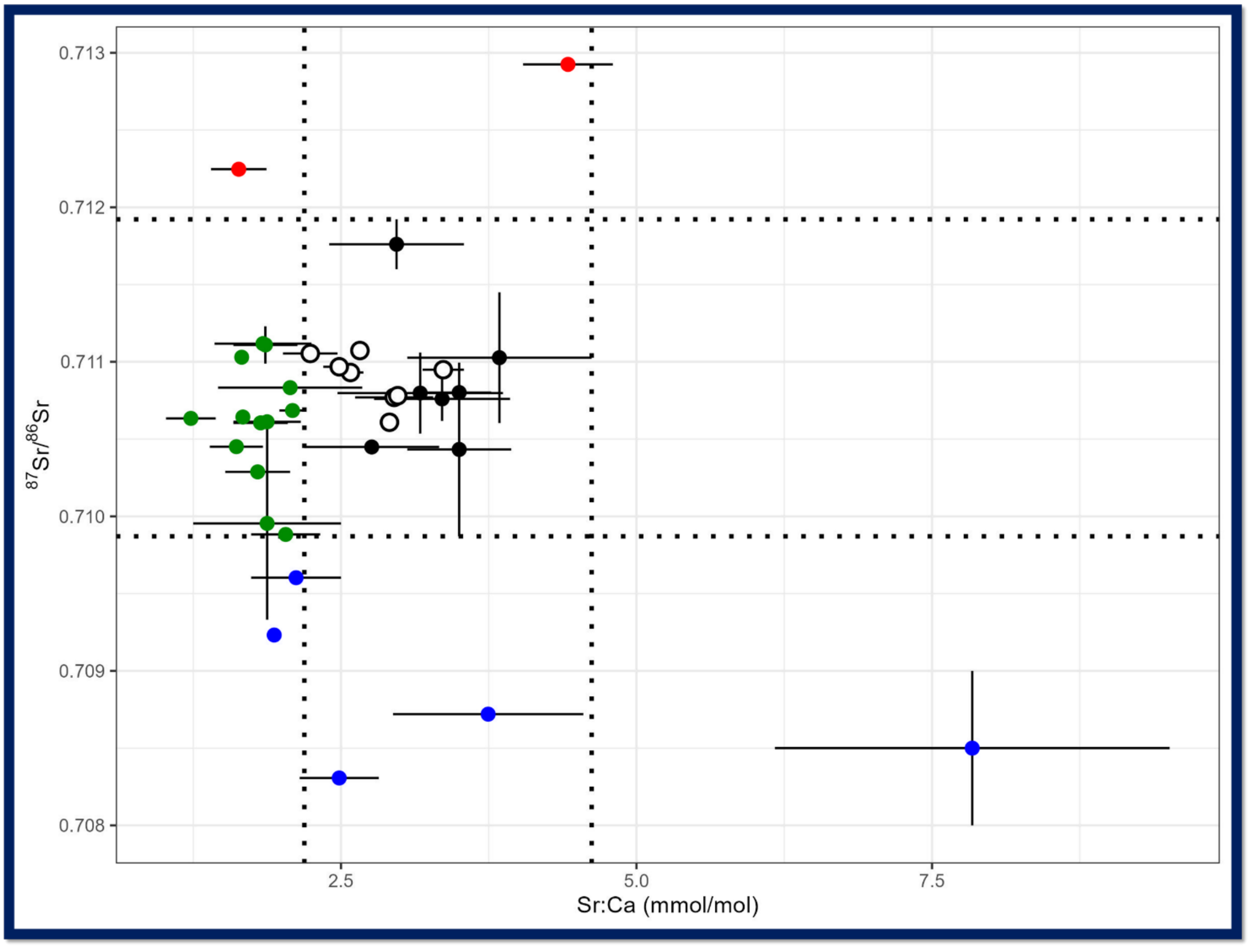

Figure 13. Water strontium isotope ratio ($^{87}Sr/^{86}Sr$) and strontium:calcium ratio (Sr:Ca) for Ohio River navigation pools (black dots) and tributaries. Horizontal and vertical bars around data points indicate observed ranges (including data from this study and prior studies) of Sr:Ca and $^{87}Sr/^{86}Sr$, respectively. Dashed lines indicate minimum and maximum Sr:Ca and $^{87}Sr/^{86}Sr$ values across all Ohio River sites. Red dots indicate tributaries with higher $^{87}Sr/^{86}Sr$ than the Ohio River, blue dots represent tributaries with lower $^{87}Sr/^{86}Sr$ than the Ohio River, green dots indicate tributaries within the range of Ohio River $^{87}Sr/^{86}Sr$ values but with lower Sr:Ca than the Ohio River, and hollow (white) dots indicate tributaries that cannot be distinguished from the Ohio River using either Sr:Ca or $^{87}Sr/^{86}Sr$.

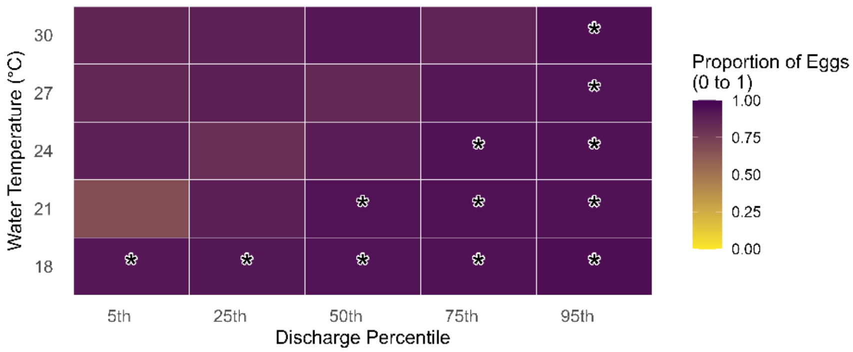

Figure 14. Predicted proportion of viable Silver Carp eggs spawned in the Great Miami River across different combinations of discharge and water temperature scenarios. Asterisks indicate scenarios in which viable eggs were predicted to drift out of the Great Miami River and into the Ohio River mainstem.

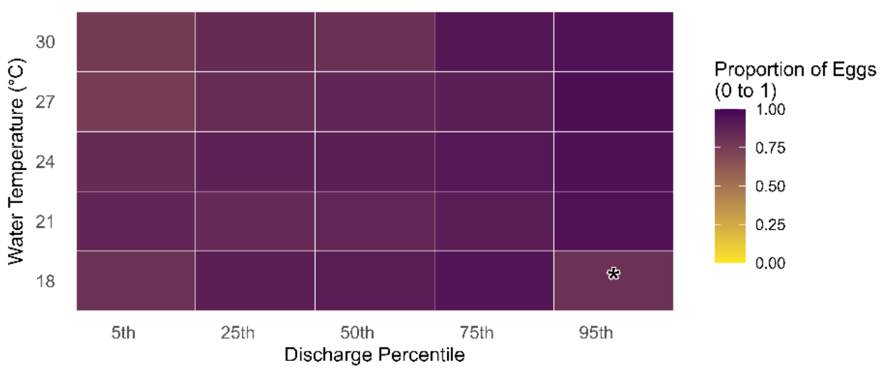

Figure 15. Predicted proportion of viable Silver Carp eggs spawned in the Scioto River across different combinations of discharge and water temperature scenarios. Asterisks indicate scenarios in which viable eggs were predicted to drift out of the Scioto River and into the Ohio River mainstem.

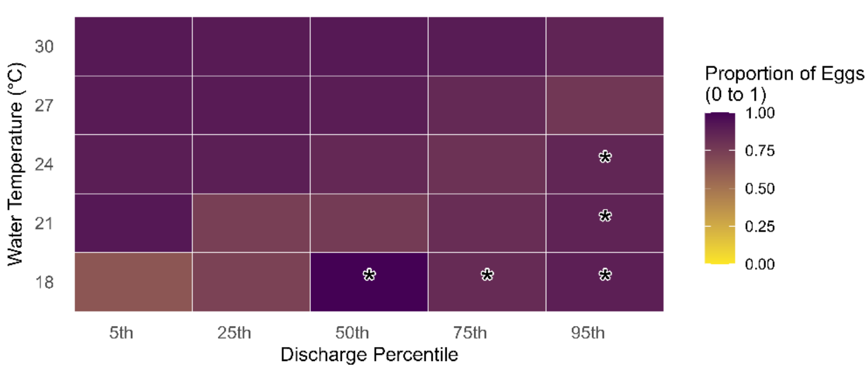

Figure 16. Predicted proportion of viable Silver Carp eggs spawned in the Hocking River across different combinations of discharge and water temperature scenarios. Asterisks indicate scenarios in which viable eggs were predicted to drift out of the Hocking River and into the Ohio River mainstem.

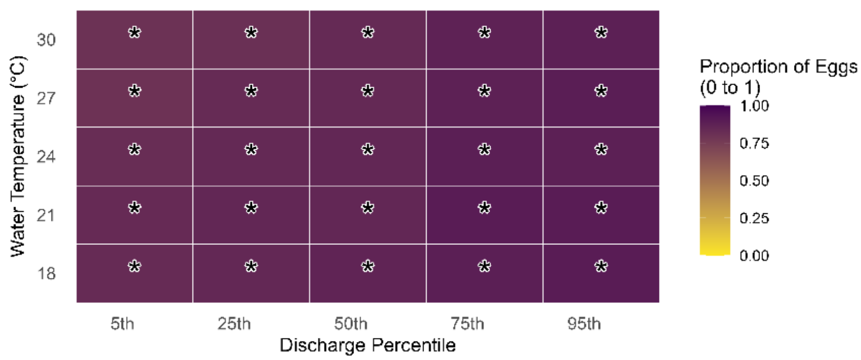

Figure 17. Predicted proportion of viable Silver Carp eggs spawned in the Muskingum River across different combinations of discharge and water temperature scenarios. Asterisks indicate scenarios in which viable eggs were predicted to drift out of the Muskingum River and into the Ohio River mainstem

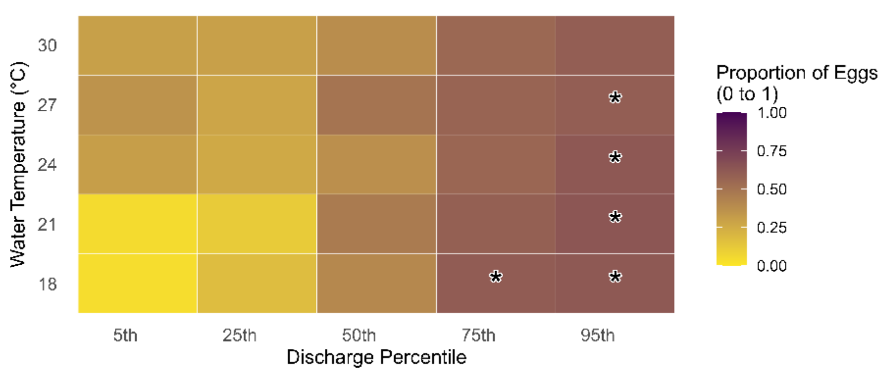

Figure 18. Predicted proportion of viable Silver Carp eggs spawned in the Little Miami River across different combinations of discharge and water temperature scenarios. Asterisks indicate scenarios in which viable eggs were predicted to drift out of the Little Miami River and into the Ohio River mainstem

Recommendations:

- Continue standardized larval sampling throughout the Ohio River Basin to quantify density of invasive carp eggs and larvae for evaluating contribution from different river reaches. Use this information to monitor changes in spawning success over time.
- We recommend the continued use of morphometric methodologies being paired with genetic confirmation of a subsample of specimens to accurately discern between invasive carp and native fish eggs and larvae. In addition, continue to improve the process of getting genetic confirmation of ichthyoplankton samples in a timely manner.
- The Meldahl Pool YOY Bighead Carp finding in 2025 highlights the need for continuing targeted sampling in Markland Pool and areas further upstream. Having multiple agencies outfitted with surface or dozer trawls will allow us to continue our expanded YOY sampling further upstream in 2026.
- We recommend moving forward with a gear comparison study in 2026 to evaluate effectiveness between the dozer trawl and surface trawl .
- KDFWR recommends continuing to develop and geographically expand invasive carp YOY surveys in the lower Ohio River basin with an emphasis on searching for YOY Black Carp. Continued effort is planned for 2026.
- We recommend additional collaboration between all project partners collecting field data to ensure that all necessary sampling information is well documented on datasheets to improve the quality of data which will allow for appropriate analysis to be performed.
- INDNR recommends continuing to monitor Bayou Drain and Hovey Lake to limit recruitment when possible. INDNR biologists should work closely with Hovey Lake property staff to monitor water levels and attempt to close the water control structure inlet before lake levels equalize with river level to keep YOY out.
- OSU recommends refining HEC-RAS models to more accurately represent confluence dynamics so inputs into the FluEgg modes are more representative of environmental conditions. We also recommend including more spawning sites in each tributaries crossed with discharge/temperature scenarios. Variable thermal regimes within years in contrast to constant temperatures assumed in current model runs are also recommended to improve model predictions.
- OSU recommends continuing monitoring for invasive carp in Ohio tributaries to the Ohio River near the invasion front, however, this research objective should likely be incorporated into the Early Detection and Evaluation project.

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