Title: Early detection and recovery of river herring spawning habitat use in response to a mainstem dam removal

Time period: 2015–2021

Location: Patapsco River, Maryland

Purpose: To assess the response of anadromous river herring, alewife (*Alosa pseudoharengus*) and blueback herring (*Alosa aestivalis*), to the 2018 removal of Bloede Dam by monitoring environmental DNA (eDNA), eggs, tagged fish (PIT tags), and adult fish (electrofishing samples) at locations upstream and downstream of the dam site during their spawning migrations.

Access: These data are not sensitive or classified.

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Cross reference: None

File names: Data:

eDNA\_data\_2024\_update.csv

egg\_data\_2024\_update.csv

Status: Complete

Methodology: The response of anadromous river herring, alewife (*Alosa pseudoharengus*) and blueback herring (*Alosa aestivalis*), to the 2018 removal of Bloede Dam was assessed by monitoring environmental DNA (eDNA) and eggs from 2015 to 2024 at locations upstream and downstream of the dam site during their spawning migrations.

Completeness: These data are complete.

***eDNA\_data\_2024\_update.csv***

Description:This dataset contains river herring environmental (eDNA) data collected in the Patapsco River.

File type: Comma-separated values file

Processing steps: Sampling sites were categorized into three groups according to their geographic location relative to the Bloede Dam site including “Downstream” of Bloede Dam, “Restored” sites between Bloede Dam and Daniels Dam, and “Above Daniels” sites upstream of Daniels Dam. Water samples (~800 ml) were collected in autoclaved 1 L Nalgene bottles and samples were frozen in a non-defrosting freezer until further analysis. A total of 625 non-control eDNA samples were collected and processed across 16 sites from 2015 to 2023, with 159 total samples positive for river herring eDNA. All control samples (n = 55) were negative for river herring eDNA, indicating that there were no issues with sample contamination during field activities, sample storage, or sample processing. Both alewife and blueback herring eDNA was detected at sites upstream of Bloede Dam after the dam’s removal, but not before (alewife: Fig. 3A, blueback herring: Fig. 3B). Post-removal, the probability of detecting alewife eDNA at Restored sites increased from 0% to 3.4 ± 1.9%, while the probability of detecting blueback herring eDNA at Restored sites increased from 0% to 8.0 ± 2.9% (estimate ± SE, calculated using logistic regressions). In 2019, during the spring migration immediately after the removal of Bloede Dam while some construction was still ongoing, eDNA from both species was detected at the site farthest upstream in the restored segment (immediately below Daniels Dam). In addition to increased eDNA detections at Restored sites upstream of Bloede Dam, detections of eDNA increased at Downstream sites for both species. The chance of detecting alewife eDNA increased from 10.6 ± 4.0% to 30.0 ± 5.5% (odds ratio = 3.63 ± 1.69, *t*184 = 2.76, *p* = 0.006), while the chance of detecting blueback herring eDNA increased from 13.4 ± 6.4% to 36.7 ± 10.4% (odds ratio = 3.75 ± 1.70, *t*184 = 2.91, *p* = 0.004). These results provide additional evidence that river herring did not use the fish ladder on Bloede Dam but were able to move past the dam’s location immediately following removal even with some construction work continuing in spring 2019.

A river herring-specific quantitative PCR (qPCR) molecular beacon assay was used to identify river herring DNA sequences following established procedures. Briefly, water samples were thawed and filtered using 47 mm diameter Whatman cellulose nitrate filters with 1.0 μm pore size. DNA was extracted with the Omega Biotek EZNA Water kit following manufacturer’s instructions or with a CTAB Chloroform-Isoamyl extraction procedure. qPCR was conducted on sample extracts in triplicate, and samples with at least two out of three triplicates with cycle quantification (Cq) values below 39 were considered positive eDNA detections (i.e., river herring presence) (n = 402 out of 451 non-control samples). For samples with positive river herring detection, species-level identification of alewife and blueback herring was determined via Sanger sequencing. The relative ratio of alewife to blueback herring DNA in each sample was estimated based on the relative peak height ratios at a species-diagnostic SNP produced by QSVAnalyser and was used to calculate the number of eDNA copies per liter for each species. Copy numbers from qPCR amplification were then adjusted for each sample based on the filtered water volume, calculated as the number of mtDNA copies per liter of water sampled. Duplicate samples were collected from the sites from 2019 to 2021, so copy numbers were averaged across all replicates collected at the same site and time prior to analysis.

Source inputs: River herring-specific qPCR molecular beacon assay.

Entity and attributes:

Column heading: Site

Label: Site

Description: Specific location where samples were collected

Data type: Character

Measurement unit: None

Comments:

Column heading: Latitude

Label: Latitude

Description: Latitude of sampling site

Data type: Numeric

Measurement unit: Decimal degrees (DD)

Comments:

Column heading: Longitude

Label: Longitude

Description: Longitude of sampling site

Data type: Numeric

Measurement unit: Decimal degrees (DD)

Comments:

Column heading: Date

Label: Date

Description: Date of collection

Data type: Date

Measurement unit: None

Comments: Format: Month/Day/Year

Column heading: CqA

Label: CqA

Description: Cycle quantification (Cq) value of first qPCR run

Data type: Numeric

Measurement unit: qPCR cycle number

Comments: Cq values below 39 were considered positive eDNA detections (i.e., river herring presence)

Column heading: CqB

Label: CqB

Description: Cycle quantification (Cq) value of second qPCR run

Data type: Numeric

Measurement unit: qPCR cycle number

Comments: Cq values below 39 were considered positive eDNA detections (i.e., river herring presence)

Column heading: CqC

Label: CqC

Description: Cycle quantification (Cq) value of third qPCR run

Data type: Numeric

Measurement unit: qPCR cycle number

Comments: Cq values below 39 were considered positive eDNA detections (i.e., river herring presence)

Column heading: CnB

Label: CnB

Description: Copies detected in second qPCR run

Data type: Numeric

Measurement unit: Copy number from qPCR amplification

Comments:

Column heading: CnC

Label: CnC

Description: Copies detected in third qPCR run

Data type: Numeric

Measurement unit: Copy number from qPCR amplification

Comments:

Column heading: Mean\_Copies

Label: Mean Copies

Description: Mean raw copy number from three qPCR runs

Data type: Numeric

Measurement unit: Copy number from qPCR amplification

Comments:

Column heading: Detection

Label: Detection

Description: Presence or absence of river herring in sample

Data type: Numeric

Measurement unit: 0-1

Comments: 1 = present, 0 = absent

Column heading: Copies

Label: Copies

Description: Adjusted mean eDNA concentration (in copies per L)

Data type: Numeric

Measurement unit: Copies per L from qPCR amplification

Comments:

Column heading: AW\_Ratio

Label: Alewife Ratio

Description: Ratio of eDNA concentration identified as alewife

Data type: Numeric

Measurement unit: Proportion

Comments: 0-1.0

Column heading: BB\_Ratio

Label: Blueback Herring Ratio

Description: Ratio of eDNA concentration identified as blueback herring

Data type: Numeric

Measurement unit: Proportion

Comments: 0-1.0

Column heading: Year

Label: Year

Description: Year in which counts were collected

Data type: Character

Measurement unit: None

Comments:

Column heading: Removal

Label: Dam Removal

Description: Sample collection timing relative to dam removal (pre = before 2018, post = after 2018)

Data type: Character

Measurement unit: None

Comments: pre, post

Column heading: Group

Label: Group

Description: Habitat site grouping relative to Bloede Dam (Downstream, Restored, Above Daniels) for analysis

Data type: Character

Measurement unit: None

Comments: Downstream, Restored, Above Daniels

***egg\_data\_2024\_updated.csv***

Description:This dataset contains river herring egg data collected in the Patapsco River.

File type: Comma-separated values file

Processing steps: Ichthyoplankton sampling was conducted to assess spawning activity in the Patapsco River. Surveys of river herring eggs were conducted simultaneously with the collection of eDNA water samples at the “Downstream”, “Restored”, and “Above Daniels” sites following established protocols and standard methods used by Maryland DNR. A total of 466 ichthyoplankton samples were collected and processed across 18 sites from 2015 to 2024 and data for river herring eggs were analyzed for this report (the full dataset also contains data for river herring larvae). It is not possible to visually distinguish between alewife and blueback herring eggs, or between river herring and hickory shad (*Alosa mediocris*), due to morphological similarities at the early developmental stages.

Prior to statistical analysis, a qualitative lower threshold was established for the egg count data to account for potential sampling error. Observations with two or fewer eggs at any site were set as zero for “non-detection” (n = 34) and thus excluded from the average. The lower detection threshold would account for potential sampling cross-contamination, where residual eggs may not be thoroughly cleaned or removed from the net between new sampling events at different sites. Biological significance also informed the threshold, as even a single spawning female river herring can release hundreds of thousands of eggs into the water column.

Egg abundance was converted to catch per unit effort (CPUE) across the dataset, standardized as number of eggs 100 kL-1 of water. Calculating the volume of water passing through the collection net accounted for measured flow (cm s-1) at each site/sample, net area (cm2), and collection time (s). Normalized CPUE was rounded to the nearest egg to obtain an integer count value for subsequent models. Mean egg abundance was then compared among the sampling sites.

Source inputs: 46 cm x 30 cm plankton drift net with 500 μm mesh, flowmeter (JDC Electronics Flowatch)

Entity and attributes:

Column heading: Site

Label: Site

Description: Specific location where samples were collected

Data type: Character

Measurement unit: None

Comments:

Column heading: Latitude

Label: Latitude

Description: Latitude of sampling site

Data type: Numeric

Measurement unit: Decimal degrees (DD)

Comments:

Column heading: Longitude

Label: Longitude

Description: Longitude of sampling site

Data type: Numeric

Measurement unit: Decimal degrees (DD)

Comments:

Column heading: Date

Label: Date

Description: Date of collection

Data type: Date

Measurement unit: None

Comments: Format: Month/Day/Year

Column heading: Herring\_Eggs

Label: Herring Eggs

Description: Number of river herring eggs collected in sample

Data type: Numeric

Measurement unit: Count

Comments:

Column heading: Year

Label: Year

Description: Year in which counts were collected

Data type: Number

Measurement unit: None

Comments:

Column heading: Month

Label: Month

Description: Month in which counts were collected

Data type: Number

Measurement unit: None

Comments:

Column heading: Day

Label: Day

Description: Day of the month on which counts were collected

Data type: Number

Measurement unit: None

Comments:

Column heading: Group

Label: Group

Description: Habitat site grouping relative to Bloede Dam (Downstream, Restored, Above Daniels) for analysis

Data type: Character

Measurement unit: None

Comments: Downstream, Restored, Above Daniels

Column heading: Removal

Label: Dam Removal

Description: Sample collection timing relative to dam removal (pre = before 2018, post = after 2018)

Data type: Character

Measurement unit: None

Comments: pre, post

Column heading: Herring\_Eggs\_Threshold

Label: Herring Eggs Threshold

Description: Number of river herring eggs in sample adjusted for detection threshold (if Herring\_Eggs < 3, set to zero as non-detection)

Data type: Numeric

Measurement unit: Count

Comments:

Column heading: PA

Label: Presence-absence

Description: Presence or absence of river herring eggs in sample

Data type: Number

Measurement unit: None

Comments: 1 = present, 0 = absent

Column heading: Flow

Label: Flow

Description: Measured stream flow (in cm per second)

Data type: Numeric

Measurement unit: Centimeters per second (cm/s)

Comments:

Column heading: Sample\_Time

Label: Sample Time

Description: Amount of time stationary net was deployed for sample collection (in seconds)

Data type: Numeric

Measurement unit: Seconds

Comments:

Column heading: Volume

Label: Volume

Description: Volume of water sampled for eggs calculated by accounting for measured flow, net area, and collection time.

Data type: Numeric

Measurement unit: Kiloliters (kL)

Comments:

Column heading: EggCPUE

Label: Egg CPUE

Description: Normalized egg count per unit effort, by dividing Herring\_Eggs\_Threshold by Volume.

Data type: Numeric

Measurement unit: Eggs per kL

Comments:

Column heading: adj\_EggCPUE

Label: Adjusted Egg CPUE

Description: Normalized egg CPUE, rounded to the nearest egg (number of eggs per 100 kL of water)

Data type: Numeric

Measurement unit: Eggs per 100 kL

Comments:

Column heading: log\_adj\_EggCPUE

Label: Log Adjusted Egg CPUE

Description: Log transformed normalized egg CPUE

Data type: Numeric

Measurement unit: Log(Eggs/100 kL)

Comments: