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Determining the Effects of Landlocked Alewives on Anadromous Alewife Restoration

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Abstract

Dam removal and fish passage projects are a critical component of anadromous alewife restoration, reconnecting runs to prime spawning habitat in coastal lakes. However, landlocked alewife populations have become established in many coastal New England lakes. The effects of landlocked alewives on anadromous alewife restoration are currently unknown. We investigated the effects of landlocked alewife presence on anadromous alewife restoration in Rogers Lake, which once hosted one of the largest anadromous alewife runs in Connecticut. Thus, effective restoration could substantially bolster regional alewife production. From 2015-2017, we stocked spawning anadromous adults into Rogers Lake. Each summer, we sampled juvenile alewives from the lake. We developed a novel set of microhaplotype genetic markers to identify anadromous, landlocked, and hybrid juveniles. Estimates of spawning time show that anadromous alewives spawn earlier in the spring than landlocked alewives, but that there is a period of overlap in spawning time. Therefore, the potential for hybridization between life history forms does exist. We genotyped a total of 3,171 alewife specimens representing landlocked fish prior to stocking, stocked anadromous adults, and juveniles collected from the lake after the initiation of anadromous stocking. Results of genetic monitoring indicate that anadromous alewives are successfully spawning in Rogers Lake. From our sample of juveniles genotyped in 2017, we identified 1,154 landlocked individuals (90.6%), 75 anadromous individuals (5.9%), and 45 hybrids (3.5%). The identification of anadromous juveniles indicates that anadromous alewife are able to successfully spawn and juveniles to rear in a lake containing a landlocked population. The identification of hybrids indicates that the two life history forms can successfully spawn together and produce viable and competitive offspring. Applying our proportions to the total number of juvenile alewives in Rogers Lake, we estimate that the lake contained about 400,000 landlocked, 26,000 anadromous, and 15,500 hybrid juveniles in August 2017. These estimates suggest that anadromous production is high enough to initiate anadromous alewife restoration. They also show that landlocked alewives are still substantially more common in the lake compared to anadromous or hybrids. Hybrids are less common than anadromous juveniles, but they are present at ecologically and evolutionarily relevant abundances. Future work will continue to track the abundance of each life history form to better understand how anadromous production and hybridization are proceeding as the restoration project continues.

1. Stocking and sample collection

Starting in 2015, we undertook a study examining the reintroduction of anadromous alewives to Rogers Lake, Connecticut. This project was the culmination of a decades-long effort to restore anadromous alewife access to the lake. Dams constructed before 1800 blocked access to this lake from anadromous alewives, leading to the evolution of a landlocked alewife population in the lake (Palkovacs et al. 2008, Twining and Post 2013). The effects that this resident landlocked alewife population would have on anadromous alewife restoration were not known. Our goals were to **1)** use otolith-based estimates of spawn timing to evaluate the potential for hybridization between anadromous and landlocked alewives, **2)** use novel genetic markers to identify successful anadromous spawning and identify instances of hybridization, and **3)** use juvenile density estimates to determine the abundance of landlocked, anadromous, and hybrid alewives in Rogers Lake.

Our initial goal was to allow anadromous alewife re-introduction to proceed via the opening of the Rogers Lake fishway in 2015. Unfortunately, the anadromous alewife run in Mill Brook, the outlet stream from Rogers Lake, has declined to such low abundances that no fish successfully made it to the lake on their own. As an emergency measure, we collected spawning adults from downstream in Mill Brook late in the spawning season and moved them to the lake via stocking truck. While this method is logistically challenging, it has the advantage of allowing us to collect fin clips for genotyping from all anadromous adults stocked into the lake. Because of the decline of the Mill Brook run, we decided to introduce adult anadromous spawners in 2016 and 2017 from the nearby Bride Brook run. The Bride Brook run is one of the few remaining strong alewife runs in Southern New England, and genetic analysis revealed that this run is not significantly differentiated from the Mill Brook run. Thus, it is an appropriate source for stocking Rogers Lake. The numbers of spawning adult alewives stocked into Rogers Lake over the study period are given in Table 1. We anticipated that 500-1000 adults would enter the lake each year during the duration of this study. While the 2015 number fell short due to the lack of fish at Mill Brook, the numbers stocked in 2016 and 2017 from Bride Brook allowed us to exceed our expectation. Sex, length, and fin clips for genetic analysis were obtained from all adults introduced into the lake.

Table 1: Anadromous adults stocked into Rogers Lake

Year	Activity	Total	Female	Male
2015	Adults stocked into Rogers from Mill Brook	134	37	97
2016	Adults stocked into Rogers from Bride Brook	1144	713	431
2017	Adults stocked into Rogers from Bride Brook	2787	1166	1621

Each August, we collected juvenile alewives from Rogers Lake using a pelagic purse seine. The seine (composed of 3.18-mm mesh) is 4.87 m deep and 35.36 m long and encircles an area of 100 m^2. Our goal was to collect 1,000 juvenile alewives from the lake each year for genetic analysis. This sample size is necessary to detect hybridization at rates <1%. Such low levels of hybridization may be likely when anadromous alewives first recolonize the lake in relatively small numbers. The numbers of juvenile alewives collected from Rogers Lake over the study period are given in Table 2. We exceeded our sampling goal in two of the three years of the study, allowing us to robustly estimate the proportions of anadromous, landlocked, and hybrid juveniles in the lake in 2016 and 2017. Length, weight, and fin clips for genetic analysis were obtained from all juveniles collected.

Table 2: Juvenile alewives collected from Rogers Lake each August

Year	Activity	Total
2015	Juveniles collected in Rogers Lake for genetic monitoring	450
2016	Juveniles collected in Rogers Lake for genetic monitoring	1120
2017	Juveniles collected in Rogers Lake for genetic monitoring	1410

2. Spawning Time Overlap

Overlap in spawning time is one main factor that may limit the opportunity for hybridization between landlocked and anadromous alewife populations. Using samples collected from 2014-2015, we estimated the distribution of spawning time for landlocked alewives from Rogers Lake and anadromous alewives from Bride Brook. We used otolith daily growth increments to back-calculate age. Spawning date was back calculated by combining age with temperature-dependent estimates of development time within the eggs (Littrell et al. In review).

Our results indicated that anadromous alewife spawned earlier and over a shorter duration than landlocked alewife. In 2014 and 2015, the mean spawn date for anadromous alewives occurred on May 19, whereas mean spawn data for landlocked alewives occurred on June 29. Nonetheless, there was a 3% overlap in the distributions of spawning time (overlap in late May - early June), indicating the potential for hybridization between landlocked and anadromous alewife in Rogers Lake. The degree of spawning time overlap was primarily driven by yearly variation in landlocked alewife spawning time, whereas the timing and duration of spawning for anadromous alewife runs was found to be stable across years (Littrell et al. In review).

3. Genetic Monitoring

We developed a novel set of 'microhaplotype' genetic markers for specific use in this project. Microhaplotypes are short DNA fragments containing multiple single nucleotide polymorphisms (SNPs). Each microhaplotype contains 2 – 5 SNPs per fragment, which in turn leads to more information per locus. The major technical advantages of this technique are more information per locus and the ability to account for any new variants detected (which is not possible with the SNP assays). Power analysis confirmed the reliability of these markers to distinguish landlocked, anadromous, and hybrid individuals with very high confidence.

We have completed microhaplotype genotyping of a subset of the anadromous adults stocked into Rogers Lake and juveniles collected from the lake in 2017. The numbers of samples genotyped are given in Table 3. We focused our effort on genotyping adults and juveniles from 2017, since the large number of anadromous adults stocked in this year gave us the best chance of finding anadromous spawning and hybridization.

Table 3: Number of samples genotyped

Year	Description	Number Genotyped
2013	Rogers Lake landlocked before stocking	122
2014	Rogers Lake landlocked before stocking	73
2015	Anadromous adults (stocked from Mill Brook)	134
2016	Anadromous adults (stocked from Bride Brook)	48
2017	Anadromous adults (stocked from Bride Brook)	1,520
2017	Juveniles collected from Rogers Lake	1,274
	Total Fish Genotyped	3,171

We identified anadromous, landlocked, and hybrid individuals using the Bayesian clustering method implemented in the program NewHybrids (Anderson and Thompson 2002). From our sample of 1,274 juveniles genotyped in 2017, we identified 1,154 landlocked individuals (90.6%), 75 pure anadromous individuals (5.9%), and 45 hybrids (3.5%).

4. Juvenile density estimates

From 2013-2017, we sampled juvenile alewife density in Rogers Lake (sampling conducted between August 5-15 of each year) using replicated purse seine samples (Table 4). Each sample consisted of 6-9 purse seine sets, where three sets were conducted in the littoral zone and the remainder were collected in the pelagic zone of the lake. Habitat-specific density estimates were multiplied by the area of Rogers Lake represented by each habitat type to arrive at an estimate of the total number of juvenile alewife in Rogers Lake.

Table 4: Juvenile alewife density estimates for Rogers Lake

Year	Purse seine sets	Alewife density per 100 m^2 (SD)	Rogers Lake juvenile density estimate (SD)
2013	6	19.00 (8.86)	201,400 (93,949)
2014	7	12.04 (4.51)	127,642 (47,765)
2015	9	40.67 (22.47)	431,067 (238,132)
2016	6	9.33 (3.48)	98,933 (40,748)
2017	6	41.67 (16.83)	441,667 (178,355)

We applied the proportions of each alewife type (landlocked, anadromous, hybrid) from our sample of genetic identifications to the total number of juveniles estimated in 2017 to arrive at estimates of the total number of each alewife life history form in Rogers Lake during August 2017. We estimate that Rogers Lake contained:

- Landlocked: 400,150 (95% CI: 217,040 - 582,940)
- Anadromous: 26,058 (95% CI: 14,130 - 37,960)
- Hybrids: 15,458 (95% CI: 8,385 - 22,520)

These estimates suggest that anadromous production is high enough to initiate anadromous alewife restoration. They also show that landlocked alewives are still substantially more common in the lake compared to anadromous or hybrids. Hybrids appear less common than anadromous juveniles (although the 95% confidence intervals do overlap), but they are present at ecologically and evolutionarily relevant abundances. Future work will continue to track the abundance of each life history form to better understand how anadromous production and hybridization are proceeding as the restoration project continues.

5. Relation of this project to the SWG award

The SWG and RCN awards are complimentary in terms of allowing this work to be completed. The SWG award supported the spring anadromous adult stocking efforts and associated tissue collections, annual summer juvenile sampling and tissue collections, and otolith data collection for estimating spawning times.

The RCN award supported the development of new genetic tools, collection of genotype data, and genetic identification of anadromous, landlocked, and hybrid alewife from Rogers Lake.

6. References

Anderson, E. C. and E. A. Thompson. 2002. A model-based method for identifying species hybrids using multilocus genetic data Genetics 160:1217–1229.

Littrell, K. A., D. Ellis, S. R. Gephard, A. D. MacDonald, E. P. Palkovacs, K. Scranton, and D. M. Post. In review. Evaluating the potential for pre-zygotic isolation and hybridization between landlocked and anadromous alewife (*Alosa pseudoharengus*) following secondary contact. Evolutionary Applications.

Palkovacs, E. P., K. B. Dion, D. M. Post, and A. Caccone. 2008. Independent evolutionary origins of landlocked alewife populations and rapid parallel evolution of phenotypic traits. Molecular Ecology 17:582-597.

Twining, C. W. and D. M. Post. 2013. Cladoceran remains reveal presence of a keystone size-selective planktivore. Journal of Paleolimnology 49:253-266.

7. Products

This award produced two papers (both accepted pending minor revisions), one currently in preparation, and several others planned. We gave one oral presentation (at the Northeast Fish and Wildlife Conference in 2017) and have others planned, including an abstract submitted to the American Fisheries Society Annual Meeting in 2018. We have also participated in a wide range of outreach and public education events.

In addition, this award contributed to the training of several junior scientists, including two PhD students (Katherine Littrell, Yale University; Diana Baetscher, UC Santa Cruz) and one postdoctoral researcher (Kerry Reid, UC Santa Cruz).

Papers

Reid, K., E. P. Palkovacs, D. J. Hasselman, D. Baetscher, J. Kibele, P. Bentzen, M. C. McBride, and J. C. Garza. In review. Comprehensive evaluation of genetic population structure for anadromous river herring with single nucleotide polymorphism data. Fisheries Research.

Littrell, K. A., D. Ellis, S. R. Gephard, A. D. MacDonald, E. P. Palkovacs, K. Scranton, and D. M. Post. In review. Evaluating the potential for pre-zygotic isolation and hybridization between landlocked and anadromous alewife (*Alosa pseudoharengus*) following secondary contact. Evolutionary Applications.

Talks

Post, DM; Garza, JC; Gephard, S; Littrell, K.; Reid, K; Palkovacs, EP. 2017. Determining the Effects of Landlocked Alewives on Anadromous Alewife Restoration. 73rd Annual Northeast Fish and Wildlife Conference (April 11, 2017; Norfolk, VA).

Poster Presentations

Reid K, Garza JC, Post D, Palkovacs EP. 2018. Tracking the outcome of secondary contact between fish lineages in a whole-lake experiment. 2018. University of California Santa Cruz Postdoc Symposium (March 1, 2018; Santa Cruz, CA).

Outreach

Ellis, D. 2016. Tour of the Bride Lake Alewife Trap, for agency staff. East Lyme, CT. (April 22, 2016).

Ellis, D. 2016. Tour of the Bride Lake Alewife Trap, for University of Connecticut class. East Lyme, CT. (April 8, 2017).

Gephard, S. 2016. Talk to public on River Herring. Sponsored by Connecticut River Watershed Council, East Hartford, CT. (February 23, 2016).

Gephard, S. 2016. River herring volunteer training orientation. Presentation made to educate and train observers of river herring runs for data collection. Connecticut River Watershed Council, Middletown, CT. (March 24, 2016).

Gephard, S. 2016. Interview about river herring restoration on WTIC radio, 1080-AM Hartford. (April 7, 2016).

Gephard, S. 2016. River Herring Research and Conservation. Subject of weekly half-hour radio program devoted to migratory fish. iCRV on-line streaming radio. http://icrvradio.com/. Essex, CT. (April 27, 2016).

Gephard, S. 2017. Testimony to the New England Fisheries Management Council on the need to close areas in offshore federal waters to the Atlantic Herring Fishery for the conservation of river herring. NEFMC, Mystic, CT. (April 19, 2017).

Gephard, S. 2017. River Herring Research and Conservation. Subject of weekly half-hour radio program devoted to migratory fish. iCRV on-line streaming radio. http://icrvradio.com/. Essex, CT. (May 24, 2017).

Gephard, S. 2017. Testimony to the New England Fisheries Management Council on the need to close areas in offshore federal waters to the Atlantic Herring Fishery for the conservation of river herring. NEFMC, Newport, RI. (December 5, 2017).

Gephard, S. 2017. Presentation to Connecticut Fisheries Advisory Council on the proposed Amendment 8 of the Atlantic Herring Fisheries Management Plan of the New England Fisheries Management Council—pertaining to the conservation of river herring. Hartford, CT. (December 6, 2017).

Palkovacs, E.P. 2016. Testimony to the Mid-Atlantic Fishery Management Council on the proposed inclusion of river herring under Federal management. (October 6, 2016; Galloway, NJ).

Palkovacs, E.P. 2018. River herring population genetic structure and genetic stock identification of bycatch. Presentation to the Northeast Inter-agency Regional River Herring Meeting. (February 6, 2018).

Palkovacs, E.P. 2018. River herring population genetic structure and genetic stock identification of bycatch. Presentation to the River Herring Stock Status Review Team. (March 1, 2018).