FINAL REPORT

State: **NEW HAMPSHIRE** **Grant**: **F-61-R-25/F22AF00514**

Grant Title: **NEW HAMPSHIRE'S MARINE FISHERIES INVESTIGATIONS**

Project I: **DIADROMOUS FISH INVESTIGATIONS**

Job 4: **MONITORING OF NEW HAMPSHIRE RAINBOW SMELT RESOURCES**

Objective: To release hatchery-reared, oxytetracycline-marked Rainbow Smelt *Osmerus mordax* for population enhancement and assess potential contributions and among-river movements using passive integrative transponders and otolith analyses.

Period Covered: January 1, 2019 - December 31, 2023

ABSTRACT

Between January 1, 2019 and December 31, 2023, the New Hampshire Fish and Game Department worked in collaboration with the University of New Hampshire to conduct one investigation per year regarding stock enhancement and additional monitoring of rainbow smelt movements and survival during their spawning run in the Great Bay Estuary. During 2019-2021, the New Hampshire Fish and Game Department, in collaboration within the University of New Hampshire (UNH), released a total of 7,172,500 Rainbow Smelt (*Osmerus mordax*) larvae into Great Bay for stock enhancement purposes. Released rainbow smelt were first bathed in oxytetracycline to mark otoliths for future identification. Released larvae came from two sources: 1) the Aquaculture Research Center (ARC) at UNH, and 2) imported from Harmon Brook Smelt Hatchery. To assess potential contributions of released larvae to adult populations, adult rainbow smelt (age 1+) were collected on spawning grounds during annual fyke net surveys and their otoliths sectioned, polished, and read for potential oxytetracycline marks. Most of these efforts occurred in 2022, with 165 otoliths read, with no evidence of oxytetracycline marks, making it difficult to quantify potential contributions of stock enhancement. Also in 2022, a passive integrative transponder (PIT) tag study was initiated to describe rainbow smelt movements and survival. In 2022, 131 Rainbow Smelt were given PIT tags, and an additional 545 Rainbow

Smelt were given PIT tags in 2023. Of these 545 Rainbow Smelt PIT-tagged, 423 of them were encountered on the Winnicut River, and across rivers 503 were male (~92%). Although recaptures occurred within season (generally on the same river as originally tagged), no fish tagged in 2022 were recaptured during 2023 efforts, indicated low recapture rates.

INTRODUCTION

Anadromous Rainbow Smelt *Osmerus mordax* are an important recreational fish in New Hampshire and were of high commercial value, historically. They are small anadromous fish that live in nearshore coastal waters and spawn in the spring in tidal rivers immediately above the head of tide in freshwater (Kendall 1926; Murawski et al. 1980; Buckley 1989). Anadromous smelt serve as important prey for recreational, commercial, and culturally valuable species, such as Atlantic Cod *Gadus morhua*, Atlantic Salmon *Salmo salar*, and Striped Bass *Morone saxatilis* (Clayton et al. 1978; Kircheis and Stanley 1981; Stewart et al. 1981; Kirn 1986; O'Gorman et al. 1987). The range of smelt historically extended from Chesapeake Bay to Labrador (Kendall 1926; Buckley 1989); but over the last century, the range has contracted and smelt are now only found east of Long Island Sound, and recent studies suggest it may only extend as far south as Buzzards Bay, Massachusetts (Enterline et al. 2012).

Rainbow Smelt have supported culturally important commercial and recreational fisheries throughout New England since at least the 1800s. Concerns have risen about the population status of smelt in recent years. High numbers of smelt that once supported commercial fisheries in New England have declined precipitously since the late 1800s to mid-1900s (Enterline et al. 2012). The current status of Rainbow Smelt populations for the majority of the Gulf of Maine is not well known. There has been a 15-20 year decline in the Massachusetts Bay region smelt populations. New Hampshire's commercial landings of Rainbow Smelt, once as high as 110,000 pounds in the mid-1960's, declined to less than 30,000 pounds by the mid-1970's, and rapidly declined from there.

The National Oceanic and Atmospheric Administration listed Rainbow Smelt as a federal Species of Concern in 2004 because of over-harvest, water quality and habitat degradation, inaccessibility to spawning grounds, and possible disease issues. New Hampshire also lists anadromous smelt as a Species of Special Concern. Although smelt population declines have been widely documented, the causes are not well understood. In the federal listing of smelt, factors identified as potential contributors included structural impediments to their spawning migration (such as dams and blocked culverts) and

chronic degradation of spawning habitat due to storm water inputs that include toxic contaminants, nutrients, and sediment (Chase and Childs 2001).

Further evidence of the decline of Rainbow Smelt can be derived from a survey of historically active spawning sites throughout ME, using a study from the 1970s (Flagg 1974) as a valuable baseline for comparison. A recent survey found that 13% of the historically active spawning streams no longer support smelt spawning, and most of the streams that remain active now support smaller runs than they did historically (Enterline et al. 2012). The substantial decline in strong spawning runs warrants concern and attention throughout their range, including New Hampshire waters.

The New Hampshire Fish and Game Department (NHFG) monitors the Rainbow Smelt resource in New Hampshire's Great Bay Estuary and its tributaries with a fishery-dependent creel survey in the winter months and a fishery-independent survey during the spring spawning. The winter creel survey selects for older fish in the population and fails to capture the presence of younger fish in the population, whereas age-1 fish are captured in the spring survey. Data collected from both monitoring programs are used to monitor trends in relative abundance and age, sex, and size structure of the fishery and spawning run. However, more frequent years of limited ice conditions that limit fishing opportunities may create gaps in this long-term fishery-dependent data set that may affect trend analyses in the future. Additionally, generally declining catches in the spring spawning survey may be further evidence of Rainbow Smelt population declines in New Hampshire.

In an effort to enhance population levels of Rainbow Smelt, NHFG in cooperation with the University of New Hampshire (UNH), used gravid adult smelt as broodfish to produce fry for stock enhancement from 2019-2021. These efforts ended in 2021, but these fish were OTC-marked prior to release and thus should be identifiable if they are contributing to the current adult spawning population. In response, in 2022 UNH assessed otoliths collected from rainbow smelt via existing NHFG programs to determine the proportion of stocked fish captured in the spring spawning run and juvenile finfish seine surveys. To increase ability to monitor New Hampshire Rainbow Smelt populations, UNH also implanted PIT tags into Rainbow Smelt collected by the NHFG spring spawning fyke net program in 2022 and 2023. Scanning fish on spawning grounds can be used to demonstrate within-year movements, use of multiple spawning rivers, and estimates of interannual survival. These field efforts were assisted by the Coastal Research Volunteer program (NH Sea Grant).

PROCEDURES

Rainbow Smelt were captured by fyke net (See Project I-2) in the Oyster, Squamscott, and Winnicut rivers during their spawning migrations and transported by truck to the Aquaculture Research Center (ARC) at UNH. Additionally, supplemental fry were imported from Harmon Brook Farm and Smelt Hatchery (Canaan, Maine). These fry were produced from approximately 390 female broodfish captured from the Addison River system (Washington County, Maine). All broodfish were strip spawned and the embryos incubated according to the methods of Ayer et al. (2005). Eggs were stripped into polystyrene plastic dishes by applying slight ventral pressure and fertilized with milt from 3-5 males (0.2-0.5 ml), activated with ~50 ml well water and swirled gently for 2 minutes. Fertilized eggs were transferred into a tannic acid solution (150 μL/L) and gently swirled for 10 minutes to remove adhesiveness according to the methods of Rottman et al. 1988. Fertilization success was assessed by microscopic examination 4-6 hours post-fertilization. Embryos were incubated in 5 L MacDonald hatching jars held at 10^{o}C until hatch (10-12 days post fertilization; DPF). During implementation of this project, a preliminary oxytetracycline (OTC) marking experiment was conducted on fry 1-day post hatch with concentrations of 400, 600, and 800 ppm for 4, 5, and 6-hour durations. Survival declined at concentrations ≥ 600 ppm (54%), and subsequent markings were conducted at 500 ppm for 6 hours, with survival ≥ 90%. Marking was conducted in 30 L conical tanks at 0 ppt and maintained at ~10^{o}C using ice packs. Oxytetracycline was buffered with potassium phosphate and sodium phosphate dibasic dihydrate (1:1:1 with OTC). Larvae spawned at UNH were released in the Oyster River (Jackson landing) and acclimation was accomplished by floating shipping bubbles or bags at the shoreline and slowly adding river water for 30-45 minutes. Final release was performed at the end of a pier to allow fry to drift in the current and avoid natural predators at the grassy shoreline. Larvae from Harmon Brook Farm were transported by truck to four different sites surrounding the Great Bay Estuary (Oyster, Squamscott, and Winnicut rivers, and Great Bay Proper, Fig 1.4-1). Sites were selected based on stocking truck access and to represent a variety of environmental conditions that may favor larval feeding or dispersal. Acclimation efforts were conducted on the truck by exchanging tank water with river water using buckets. Release of fry was accomplished by connecting a 3-inch diameter PVC pipe from a gate valve on the holding tank out into the current, away from the shoreline. Fry activity was observed at all release sites to monitor swimming behavior upon release.

To obtain otoliths, Rainbow Smelt are captured by fyke net (See Project

I-2) in the Oyster, Squamscott, and Winnicut rivers during spawning, when possible. Fish are sexed and measured prior to otolith extraction. Otoliths are then embedded in epoxy, sectioned, and polished to reveal growth rings and the core. Polished otolith sections were viewed under a Nikon A1R-HD CLSM at the University of New Hampshire University Instrumentation Center. Two excitation wavelengths, 409 and 487 nm, were used and detected the emitted fluorescent light with two channels if the following ranges: 435–470 nm and 500–540 nm, respectively. The resulting image sets were assigned pseudo-colors that reflected the fluorescence spectra. Volume-rendered micrographs and media files were created using FIJI. Fish and resulting otoliths were kept in opaque storage throughout all possible stages of processing and viewing to avoid potential light degradation of the mark. A total of 165 otoliths were collected from adults in 2022, with otolith collection focused on presumed age-1 fish to correspond to the ~4.1 million larval Rainbow Smelt released with oxytetracycline marks in spring 2021.

Additional Rainbow Smelt captured via fyke net (See Project I-2) on the Oyster, Squamscott, and Winnicut Rivers were given PIT tags in 2022 and 2023. PIT tags (12-mm long, Biomark) were implanted via syringe. Smelt were scanned for existing PIT tags using handheld receivers. PIT-tagged smelt were measured for fork length and total length, and scale samples were taken and aged via NHFG and UNH. Rainbow smelt collection was coordinated by NHFG and UNH with the Coastal Research Volunteer program (New Hampshire Sea Grant).

RESULTS

Between January 1, 2019 and December 31, 2023, the New Hampshire Fish and Game Department worked in collaboration with the University of New Hampshire to conduct one investigation per year regarding stock enhancement and additional monitoring of rainbow smelt movements and survival during their spawning run in the Great Bay Estuary.

Between 2019, 2020, and 2021, a total 7,172,500 larvae rainbow smelt were released into Great Bay. In 2019, broodstock were used to generate 17,500 larvae at the Aquaculture Research Center (ARC) at UNH. To supplement NHFG collection efforts, an additional 2.5 million marked larvae were imported from Harmon Brook Smelt Hatchery. In 2020, there were few sexually mature females captured on spawning grounds by NHFG, and no gravid females were transported to the ARC. Instead, approximately 2.5 million marked larvae produced using wild-caught female broodstock by Harmon Brook Hatchery were successfully released into Great Bay. In 2021, broodstock collected by NHFG

fyke net surveys were used to produce 155,000 Rainbow Smelt larvae; these were supplemented by approximately 2 million larvae from the Harmon Brook Smelt Hatchery. All larvae were marked with oxytetracycline. Across years, post-marking counts indicated survival was ≥ 90%, and mortalities were likely due to handling and transferring between containers. Larvae were released at four different spawning sites during the project period, including Oyster River (Jackson Landing), Squamscott River, Winnicut River, and Great Bay Estuary, with temperatures generally being 8-10^{o}C which closely matched marking and incubation temperatures. In 2022, concerted efforts were made to collect adult Rainbow Smelt, targeting Age-1 fish, to check otoliths for OTC marks. A total of 165 otoliths were collected, sectioned, polished, and examined. Zero otoliths showed signs of oxytetracycline marking.

PIT tagging efforts began in 2022. In 2022, a total of 131 PIT tags were deployed, with 64 on the Oyster River, 61 on the Winnicut River, and 7 on the Squamscott River. Across the three rivers, a total of 114 were male (87%), and 17 (13%) female fish were tagged. In 2023, PIT tagging efforts increased, with a total of 545 Rainbow Smelt given tags. Of these, 412 (~76%) were tagged on the Winnicut, 88 (~16%) on the Oyster, and 45 (~8%) on the Squamscott. As in 2022, the sex ratio was skewed highly towards males, with 503 males tagged (~92%) and 42 females tagged (~8%). In both years, we observed multiple within-year recapture events, generally at the same site of capture. However, no Rainbow Smelt scanned in 2023 had PIT tags deployed in 2022. All PIT-tagged Rainbow Smelt had scales sampled for aging between NHFG and UNH. Of the 131 Rainbow Smelt tagged in 2022, 101 (~77%) were age-1, 25 (~19%) were age-2, and 5 were age-3 (~4%).

DISCUSSION

For stock enhancement efforts (2019, 2020, and 2021) it was anticipated that brookstock was to be collected solely through the spring Rainbow Smelt fyke net survey each spring. However, consistently there were challenges in obtaining a high enough quantity of gravid females. From previous laboratory studies conducted at the ARC, it was estimated that to reach the target of 500,000 Rainbow Smelt larvae, it would require between 200 and 300 gravid females; roughly 2,000 larvae per female. Generally, gravid females captured in these years ranged from 0 (2020) to 8 (2019) to 41 (2021), limiting the production below the original target of 500,000 larvae per year. However, overall, these targets were exceeded annually by working with the Harmon Brook hatchery in Maine; all smelt were imported into New Hampshire via

proper permits.

In 2022, 165 otoliths of adult (mostly age-1) Rainbow Smelt collected in spring fyke net surveys were assessed for oxytetracycline, but none showed evidence of having been stocked in the system (from 2021 releases, when 2.155 million larvae were released. This lack of evidence of oxytetracycline would indicate no contributions from stock enhancement to the population, but this suggestion requires careful consideration. First, the sample size of otoliths is low, particularly given that thousands of rainbow smelt are encountered on spawning grounds each year. Second, little verification of the oxytetracycline marking was completed; this would have required rearing larvae for several months beyond ideal release points, and then checking their otoliths for valid marks. Thus, it is possible, although not probable, that not all larvae released had consistent oxytetracycline marks in their otoliths that we could observe, even if they were originally from stock enhancement efforts. If this approach were to be used in the future, efforts should be placed in rigorous validation studies prior to additional collections of otoliths from wild-caught Rainbow Smelt.

Our deployment of PIT tags reflects what was observed by NHFG Rainbow Smelt fyke net surveys on spawning grounds, which shows in 2022 and 2023 that males continue to be the dominant sex in fyke nets, with the Winnicut and Oyster Rivers having higher catch rates than the Squamscott. Several times in both 2022 and 2023 within-season recaptures occurred, implying that tags are retained and that smelt survival tag implantation. However, having zero recaptures of Rainbow Smelt in 2023 from the 2022 tags implies a very low recapture rate (< 1%). The potential causes of a low recapture rate are numerous, including 1) low tag retention, 2) low survival of tagged fish, 3) strong mixing of Rainbow Smelt among locations, not just among the three rivers monitored by NHFG, and 4) a very high population abundance of rainbow smelt (that appears unlikely). Thus, although PIT-tagging is used fairly ubiquitously among fishes with high retention and high survival, a smelt-focused study examining tagging effects would be useful in placing these results into context. In addition, to have mark-recapture efforts be effective, sample sizes should be increased. In 2023, 545 PIT tags were deployed, increasing effort more than 3-fold, increasing chances of interannual captures beginning in 2024.

PIT tag deployments largely occurred for Rainbow Smelt on the Winnicut River, males, and age-1. All of these skews generally match demographics of Rainbow Smelt observed in the fyke net surveys, with higher abundances of fish

on the Winnicut Rivers in 2022 and 2023, along with high male-to-female sex ratios. Age-1 Rainbow Smelt are the dominant age class, with ages 1-3 regularly captured; age-4+ smelt are caught more rarely. Tagging age-1 Rainbow Smelt gives the benefit of multiple potential opportunities to re-capture fish in following years (i.e., when fish are age-2 or age-3). Any recaptured fish can be also be measured (total length and fork length) to assess potential growth rates.

In conclusion, this project successfully stocked Great Bay and its tributaries with > 7 million rainbow smelt larvae over three years, but the impacts of such stock enhancement are difficult to quantify. With repeated year-after-year and increased efforts, PIT tagging may provide important insights into movements between rivers both within and among spawning seasons, and with enough sample size, estimate interannual survival. Integrating PIT tag data with ages from scales can further define age-specific patterns in habitat use and survival to help inform management strategies. Future work integrating other tagging technologies, such as acoustic telemetry (Pearson et al. In Press), can further identify important habitats used, periods of residency, and survival. Collectively these data can also be paired with other NHFG efforts (spring fyke net survey) to provide better context on cohort or age-class strength and potential interannual variability in rainbow smelt population status and productivity.

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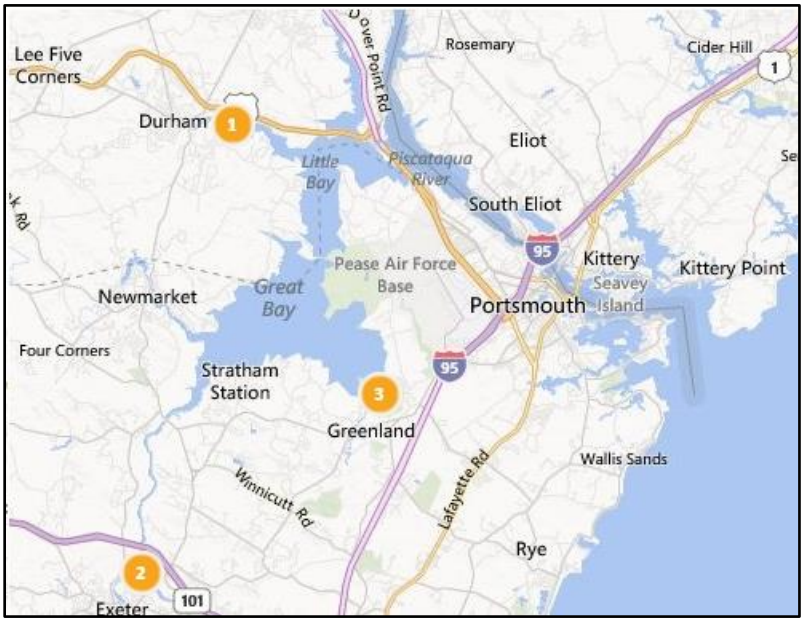
Table 1.4-2. Rainbow Smelt larvae stocking site, release date, and number of marked larvae, Great Bay Estuary, NH, 2019.

Year	Stocking site	Release date	Marked larvae
2019	Oyster River	4/4/2019	17,500
	Oyster R., Squamscott R., Great Bay Estuary and Winnicut R.	6/12/2019	2.5 million
2020	Oyster R., Squamscott R., Great Bay Estuary, and Winnicut R.	6/12/2019	2.5 million
2021	Oyster R., Squamscott R., Great Bay Estuary, and Winnicut R.	4/1/2021 6/12/2021	2.155 million

Table 1.4-2. Adult Rainbow Smelt PIT-tagged by site and sex for 2022 and 2023.

Year	River	Males tagged	Females tagged
2022	Oyster River	48	15
2022	Winnicut River	59	0
2022	Squamscott River	7	0
2023	Oyster River	83	5
2023	Winnicut River	377	35
2023	Squamscott River	43	2



1= Oyster River, 2= Squamscott River, 3= Winnicut River

Figure 1.4-1. Map of fyke net sampling locations in the Oyster, Squamscott, and Winnicut rivers, New Hampshire that were leveraged for collection of broodstock, collection of fish for otoliths, and PIT-tagging fish.

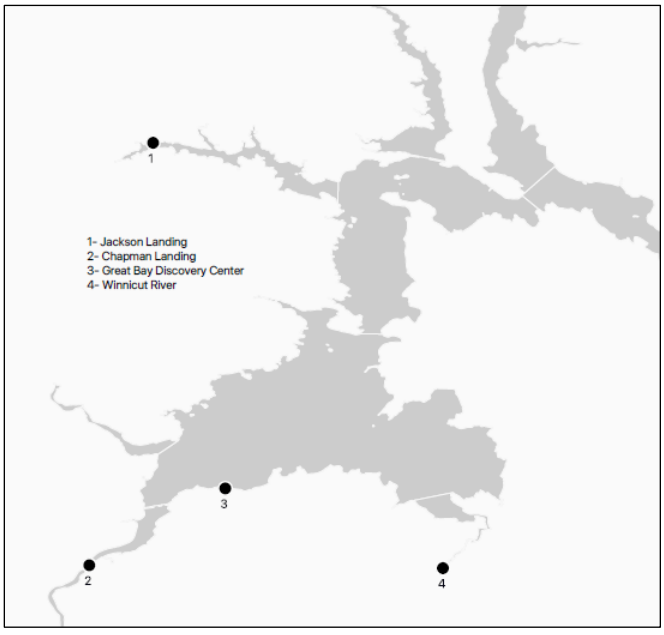


Figure 1.4-2. Location of Rainbow Smelt fry stocking sites in Great Bay estuary, NH, 2019-2021.

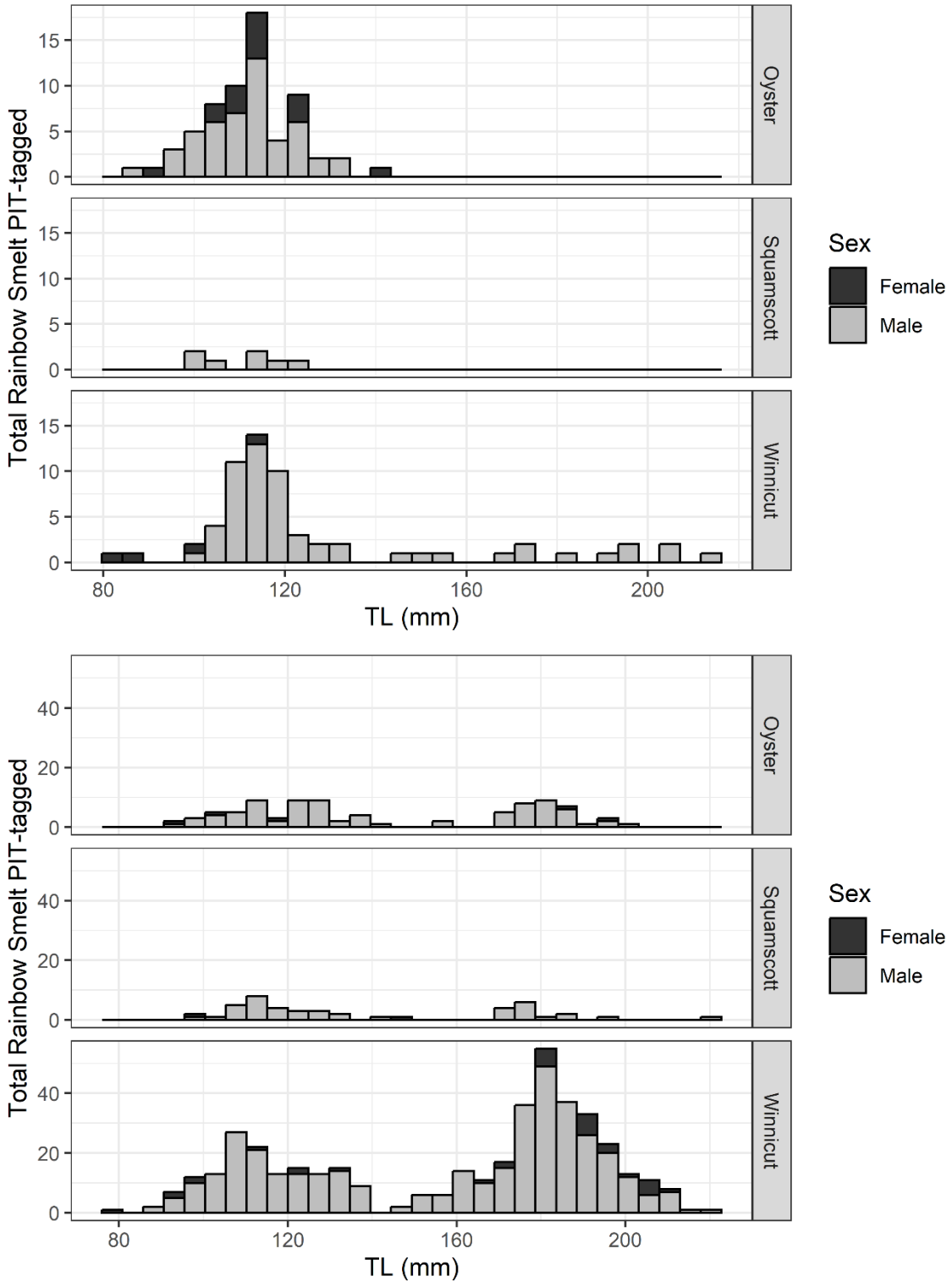


Figure 1.4-3. Total Length (TL) distributions of Rainbow Smelt receiving PIT tags among the three river systems sampled during 2022 and 2023 field seasons.