U.S. Fish and Wildlife Service
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YY Male Brook Trout Stocking and Population Monitoring in Tyee Springs 2020-2021

FY 2020-2021 Progress Report

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Disclaimers

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2020 - 2021 PROGRESS REPORT

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Abstract

Tyee Springs is located directly upstream of Carson National Fish Hatchery and is home to a thriving population of nonnative Brook Trout. This population has been a hatchery management concern and past attempts at suppressing or eradicating the Brook Trout population have been unsuccessful In 2020, the Columbia River Fish and Wildlife Conservation Office, Abernathy Fish Technology Center and Carson National Fish Hatchery initiated a long-term project using YY male Brook Trout as a tool to eradicate nonnative Brook Trout in Tyee Springs. This method involves producing male Brook Trout with two Y-chromosomes which are then released into the population targeted for eradication. Offspring of YY males (M_{yy}) and resident females (XX) are all male (XY), so over time, the population becomes skewed toward a single sex, leading to extirpation of the target population. To maximize the effectiveness of this method, annual population suppression (i.e., fish removal) is recommended prior to stocking M_{yy} Brook Trout. Population suppression and M_{yy} stocking in Tyee Springs began in 2020. We removed a total of 1,145 Brook Trout in 2020 and 2,314 in 2021, which was estimated to be 15% and 46% of the population respectively. An electrofishing tow-barge was the most effective capture method in all three habitat units, capturing 74% of total Brook Trout in 2020 and 88% in 2021. A total of 600 age-1 M_{yy} Brook Trout were stocked into Tyee Springs in 2020. All fish were adipose clipped and PIT tagged prior to release. In 2021, 1,923 age-0 M_{yy} Brook Trout were stocked into Tyee Springs. Fifty-two percent of M_{yy} individuals (999) were PIT tagged and all were marked via an adipose fin clip prior to release. Although M_{yy} Brook Trout were observed and detected with a floating PIT antenna in the following months after stocking, no M_{yy} released in 2020 were captured during the 2021 suppression event, suggesting poor overwinter survival. In contrast, five M_{yy} Brook Trout stocked in 2021 were recovered during fish removal efforts in 2022, indicating a low level of overwinter survival. Based on current fish suppression rates, sex ratios and a population model developed in 2019, we estimate females may be extirpated from the Tyee Springs population in 2033. Moving forward, we intend to continue monitoring the resident and M_{yy} Brook Trout population by analyzing tissue samples for genetic parentage analysis, tracking population sex ratios, and revising abundance estimates. These data will be used to refine the Tyee Spring population model which will in turn help assess the effectiveness of the project and the M_{yy} technique as a potential biocontrol strategy.

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Introduction

An established population of nonnative Brook Trout resides in Tyee Springs directly upstream from Carson National Fish Hatchery (NFH) in southwestern Washington. Brook Trout are one of the most prevalent nonnative fish in the western United States (Benjamin et al. 2013; Shade and Bonar 2005). Once a population becomes established, complete eradication is often difficult to achieve (e.g., Koenig et al. 2015; Meyer et al. 2006; Thompson and Rahel 1996). Traditional methods to eradicate invasive or unwanted fish include the use of fish toxicants (piscicides), targeted harvest (e.g., angling, gill nets), physical removal (e.g., electrofishing), or biological control (e.g., introduction of predators or pathogens). Tyee Springs is considered a poor candidate for piscicide treatment because of the proximity of the hatchery and presence of upwelling springs that may dilute or reduce the effectiveness of the chemical. Physical removal efforts (i.e., electrofishing) periodically implemented over nearly a decade have also proven ineffective in Tyee Springs because aquatic plant density and pockets of deep water provide refuge for fish to escape capture (USFWS 2004).

An alternative biological control strategy that could eradicate Brook Trout in Tyee Springs is the Trojan Y Chromosome approach. This method involves producing male Brook Trout with two Y-chromosomes (YY) and releasing them into the population targeted for eradication (see Shill et al. 2016). Offspring of YY males (hereafter M_{yy}) and resident females (XX) are all male (XY), so over time, the population becomes skewed toward a single sex (i.e., all males), theoretically leading to extirpation of the target population due to reproductive failure. The Idaho Department of Fish and Game has developed a broodstock of M_{yy} Brook Trout for experimental use and field stocking trials are currently underway in six western states with encouraging results (Kennedy et al. 2018; Armstrong et al. 2022).

In 2018 the Columbia River Fish and Wildlife Conservation Office (FWCO), Abernathy Fish Technology Center (FTC) and Carson NFH initiated a collaborative proof-of-concept study to assess the feasibility of stocking M_{yy} Brook Trout to eradicate nonnative Brook Trout in Tyee Springs. The outcome of this feasibility study was a population simulation model used to estimate potential time to eradication given varying levels of annual fish suppression rates and annual M_{yy} stocking rates (Poirier et al. 2020). For example, in a simulation scenario where we stock 3,000 M_{yy} Brook Trout annually and maintain a suppression rate of 50% annually (≈50% of resident Brook Trout population), our model estimates an 80% probability of eradication within 9 years. Reducing the suppression rate to 25% annually would increase time to eradication to 13 years and omitting suppression from the model completely increases this time to 19 years. These findings were consistent with previous research demonstrating that intensive annual population suppression prior to M_{yy} stocking can shorten the overall time to eradication (Schill et al. 2017; Day et al. 2020; Poirier et al. 2020; Day et al. 2021). Ultimately, we found that using the M_{yy} technique to eradicate nonnative Brook Trout in Tyee Springs was feasible and that eradication was likely to occur within a reasonable timeframe. However, achieving a 50% fish suppression rate annually would be challenging given suppression rates would decline over time as resident fish abundance decreased. A sensitivity analysis indicated no difference in time to eradication between stocking 3,000 or 7,000 Myy Brook Trout at suppression rates ≥25% (see Poirier et al. 2020). Thus, we made the decision to stock up to 3,000 M_{yy} fish annually and employ multiple fish removal methods over several months annually, to maximize suppression rates.

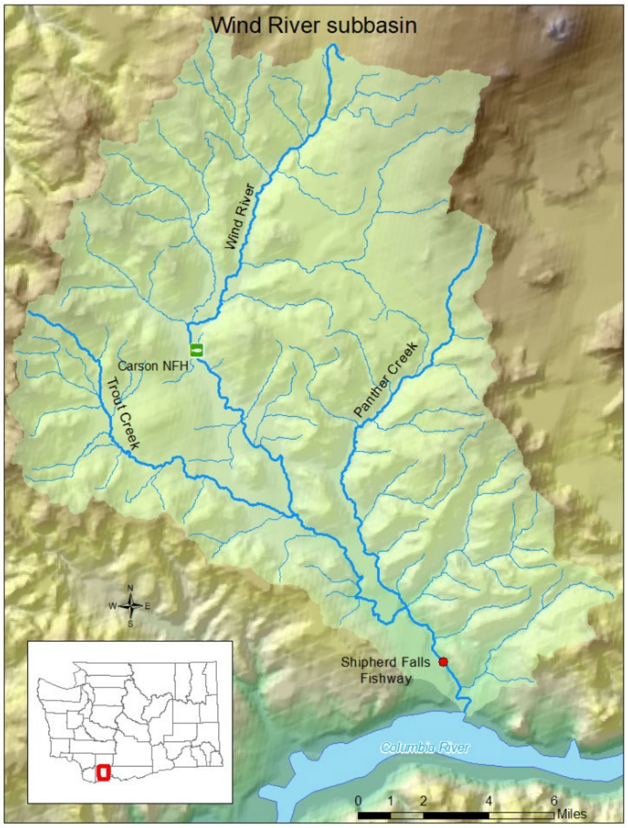
In 2020 we initiated the implementation phase of the Tyee Springs M_{yy} Brook Trout project. Our goal was to annually suppress approximately 50% of the resident Brook Trout population and stock up to 3,000 M_{yy} Brook Trout into Tyee Springs prior to the spawning season. A second important component of this project was to monitor the M_{yy} and resident Brook Trout population to assess the effectiveness of our fish removal and stocking efforts and adaptively manage implementation strategies if necessary. This report presents results of fish suppression and M_{yy} stocking efforts conducted in 2020 and 2021.

Methods

Study Area

Tyee Springs is a 0.7 km long spring-fed stream located within the Wind River watershed, approximately 19 km north of the town of Carson, Washington in Skamania County (Figure 1). Carson NFH, which began operation in 1937, was constructed to utilize Tyee Springs as its primary water source, providing year round flow at a fairly constant 6.6°C. Water from Tyee Springs enters the hatchery through an intake grate, flows through the facility and exits via the adult ladder and bypass channel before entering the Wind River at RKM 29 (Figure 1). The hatchery acts as a complete passage barrier to fish attempting to enter Tyee Springs, but a bypass channel provides some opportunity for fish to egress roughly six months of the year. Historically Carson NFH reared and released fall Chinook salmon and various trout species (including Brook Trout), before shifting to focus on spring Chinook salmon in 1981 (USFWS 2002). Although hatchery releases of Brook Trout were discontinued in 1964, a naturally reproducing population still exists in Tyee Springs. Semi-regular attempts to suppress the Brook Trout population in Tyee Springs that occurred from 1999 to 2009 did not result in the extirpation of the unwanted population (USFWS 2004). Brook Trout and sculpin are the only fish species that inhabit Tyee Springs today.

Tyee Springs is comprised of three distinct habitat segments, referred to as Units #1, #2, and #3 (Figure 1; Appendix). Unit #1 is a roughly 50 meter long, deepwater channel that starts at the hatchery intake screen at terminates at the downstream end of the Wind River road culvert. Unit #2 is the longest habitat segment and is characterized as a meandering stream stretching roughly 550 meters upstream from the Wind River road culvert to Unit #3. Unit #3 is the uppermost segment of Tyee Springs characterized as a spring-fed pond roughly 230 meters in length.



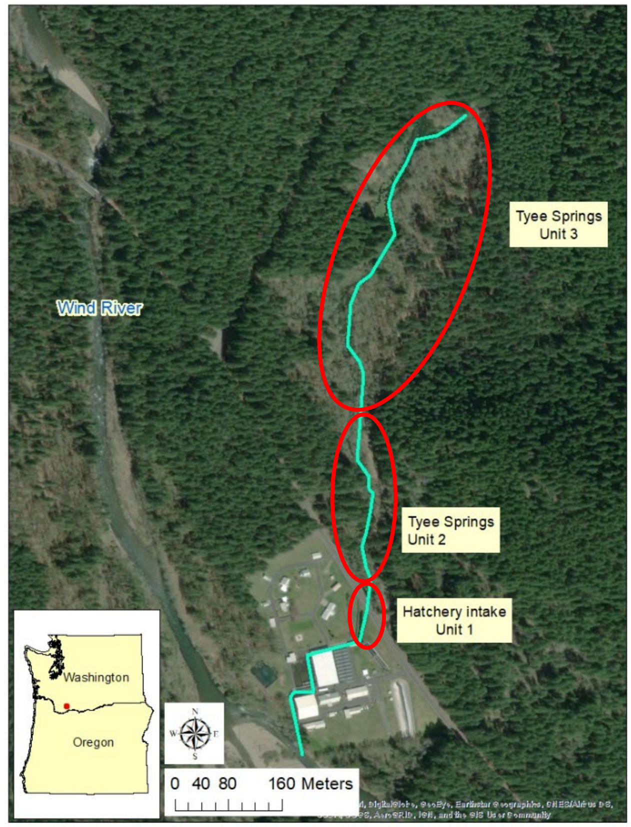


Figure 1 Map of Carson National Fish Hatchery (left) and Tyee Springs showing three habitat units (right).

Tyee Springs Population Suppression

In 2020 and 2021, Brook Trout removals occurred two days per week, during the months of July and August. We utilized up to three methods of capture that varied dependent on available staff and targeted habitat. A Smith-Root electrofishing tow-barge was utilized in all three habitat units and used exclusively in the 50 meter deepwater channel (Unit #1) immediately upstream of the hatchery intake screen. Backpack electrofishers (model LR-24 or APEX) were used in wadeable depths (<1.2 m) and backwater areas within Units #2 and #3. Minnow traps baited with salmon eggs were used briefly in 2020, but this method was abandoned after two days due to low capture rates. In 2021, we incorporated gillnets as a removal method in Unit #3 to passively capture Brook Trout overnight between consecutive sampling days. Each gillnet targeted different sized Brook Trout with 3/8 in., 1/2 in., and 3/4 in. square mesh sizes in total lengths of 50 ft., 50 ft., and 100 ft., respectively.

All Brook Trout captured during fish removal efforts were euthanized prior to sampling. In 2020, Brook Trout were euthanized in a lethal solution of tricaine methanesulfonate (MS-222), whereas in 2021 we used high concentrations of carbon dioxide (CO_2) so that captured Brook

Trout could be donated safely as a food fish. All captured fish were measured to the nearest millimeter in fork length, weighed (g), scanned for PIT tags, and inspected externally for adipose clips, deformities, wounds or parasites. A random subsample of resident Brook Trout >90 mm in length were also inspected internally to determine the sex. Approximately 100 tissue samples (i.e., fin clips) were collected from resident Brook Trout (>120 mm in length) in 2021 to establish a genetic baseline of the resident population. Tissue samples were also collected from 400 young-of-year Brook Trout in 2021 so that future genetic analyses can be used to estimate the proportion of progeny from M_{yy} parents. Collection of tissue samples was dispersed spatially and temporally throughout the field season.

M_{yy} Stocking

All M_{yy} stocked into Tyee Springs originated from Hayspur State Fish Hatchery operated by Idaho Fish and Game. Hayspur staff annually produce M_{yy} Brook Trout embryos in the fall which are subsequently transported to various projects, agencies and states for the purpose of research. Eyed Brook Trout eggs were transferred from Hayspur Hatchery to Abernathy FTC in November where they were incubated, hatched and reared until their release. Approximately one month prior to stocking, all M_{yy} were given an adipose fin clip and all (2020) or a portion (2021) of fish were implanted with a 12 mm FDX PIT tag for monitoring purposes. All M_{yy} stocked in 2020 were from brood year 2018 and fish stocked in 2021 were from brood year 2020.

Monitoring

Post-stocking mortalities

Carson NFH staff monitored hatchery intake screens for M_{yy} mortalities in the following weeks after 2020 and 2021 stocking events. Post-stocking mortalities were collected and frozen so that they could be enumerated and scanned for PIT tags at a later date.

Post-stocking PIT detections

Following M_{yy} releases, a large rectangular floating PIT antenna roughly 9-ft x 5-ft was drifted down Tyee springs to detect tagged fish and gain insight to their location, movement and survival. The floating PIT antenna was constructed by AFTC and consists of one “figure eight” style antenna (Steinke et al. 2011) attached to a foam core frame and one Biomark IS1001 to record data. The antenna was deployed in the upper most section of Unit #3 and zigzagged systematically downstream in an effort to maximize habitat coverage and repeatability.

Bypass Channel

A secondary bypass channel was constructed adjacent to the primary hatchery intake channel in 1997 to divert additional water from Tyee Springs into the Wind River during high flow events and prevent flooding of the hatchery facility. The bypass channel is typically in use for six months of the year from November through April, although timing varies annually dependent on rainfall. The bypass channel is ‘opened’ by removing dam boards, allowing surface water from Tyee Springs to drop three feet into the channel. During this time, Brook Trout residing in Tyee Springs can volitionally swim or may be flushed over the dam boards and enter the Wind River while the bypass channel is open. A PIT detection system was installed within the Carson NFH bypass channel to monitor potential Tyee Springs Brook Trout emigration to the Wind River. This system consists of one Biomark Master Controller (for multiplexing and data storage), two Biomark IS1001 readers and two antennas. Both antennas were constructed in a pass-through orientation and designed to cover 100% of the readable range within the 4-ft x 4-ft channel during peak flows (Figure 2). Antenna #1 (the upstream PIT antenna) is located roughly 20 inches downstream of dam boards that separate the bypass channel from Tyee Springs. Antenna #2 is located roughly 20 feet downstream of antenna #1.

Figure 2. FDX PIT tag antenna array located in secondary bypass channel, immediately downstream from sampling Unit #1.

Results

Tyee Springs Population Suppression

2020

Brook Trout removal efforts in 2020 fell short of expectations due to social distancing guidelines and precautions related to the worldwide COVID-19 pandemic, resulting in limited field sampling days and available personnel. Despite setbacks, 1,145 Brook Trout were removed from Tyee Springs over the course of 6 days (Table 1). The majority of Brook Trout were captured in Unit #3 with an electrofishing tow-barge (N = 901), 190 fish were removed from Unit #2 with a backpack electrofisher, and 54 fish were removed from Unit #1 with the tow-barge. Fifteen baited minnow traps were set on the first two sampling days as an experimental capture method. However, only seven Brook Trout were captured and a decision was made to discontinue minnow trapping to focus efforts on electrofishing. .

Table 1 Number of Brook Trout removed by date, habitat unit and capture method in 2020. TB = tow-barge; EF = backpack electrofisher; MT = minnow traps.

	Unit 1				Unit 2				Unit 3				
Date	**TB**	**EF**	**MT**	**Sum**	**TB**	**EF**	**MT**	**Sum**	**TB**	**EF**	**MT**	**Sum**	**Totals**
08/12/20	0	0	0	0	0	11	0	11	0	36	0	36	**47**
08/13/20	0	0	2	2	0	122	4	126	0	0	5	5	**133**
08/17/20	52	0	0	52	0	0	0	0	348	0	0	348	**400**
08/18/20	0	0	0	0	0	0	0	0	173	45	0	218	**218**
08/26/20	0	0	0	0	0	53	0	53	125	19	0	144	**197**
09/03/20	0	0	0	0	0	0	0	0	150	0	0	150	**150**
Sum	52	0	2	**54**	0	186	4	**190**	796	100	5	**901**	**1145**

Cells with zeros indicate no effort for that particular day, unit and sampling method.

The median fork length of Brook Trout removed from Tyee Springs was 112 mm, with a range of 22 to 274 mm (Figure 3). Fish captured in the deepwater channel near the hatchery intake screen (Unit #1) were larger on average than the other habitat units. The widest range of lengths were captured in Unit #3 with the electrofishing tow-barge (Figure 3). These length data were multimodal, with young-of-year fork lengths ranging from 22 mm to approximately 76 mm with a median of 53 mm. Length distributions from other age classes suffered from too much overlap to confidently identify, but likely consisted of ages one through three, based on analyses conducted in 2020 (Figure 4; Poirier et al. 2020). Field personnel attempted to determine the sex of 207 randomly selected Brook Trout during the field season (size range 87-218 mm). However, due to a lack of experience identifying the sex of small/immature individuals, we believe our error rate was high and therefore have omitted sex ratio information from analysis in 2020.

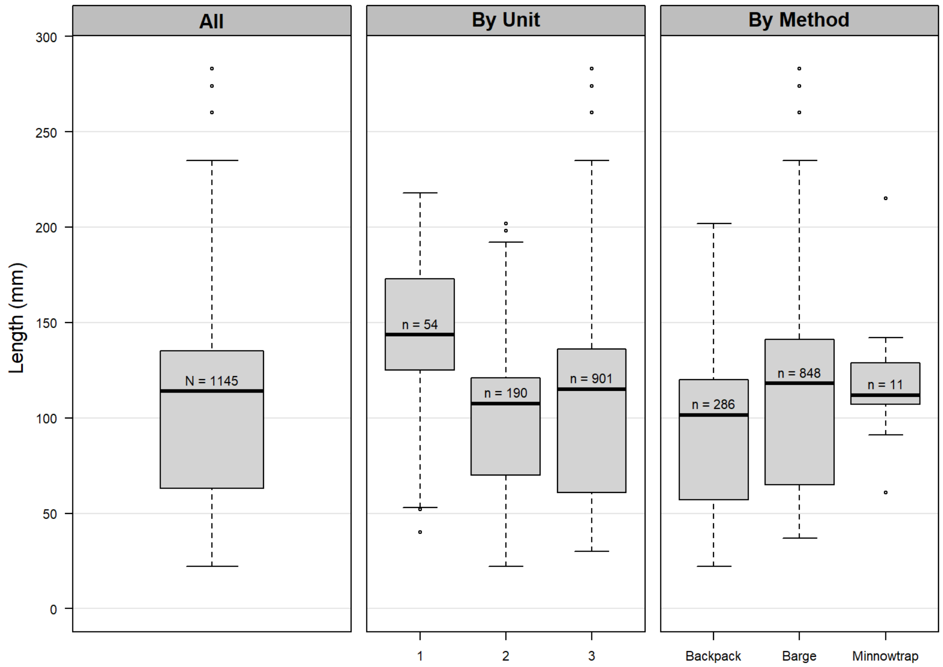


Figure 3. Boxplots of Brook Trout fork lengths from Tyee Springs removal efforts, all habitat and capture methods combined (left plot), by habitat unit (middle plot) and by capture method (right plot) in 2020. Boxes indicate the interquartile ranges (25% – 75%), whiskers indicate the total range (sans outliers), and circles indicate outliers (observations greater than 1.5 times the interquartile range).

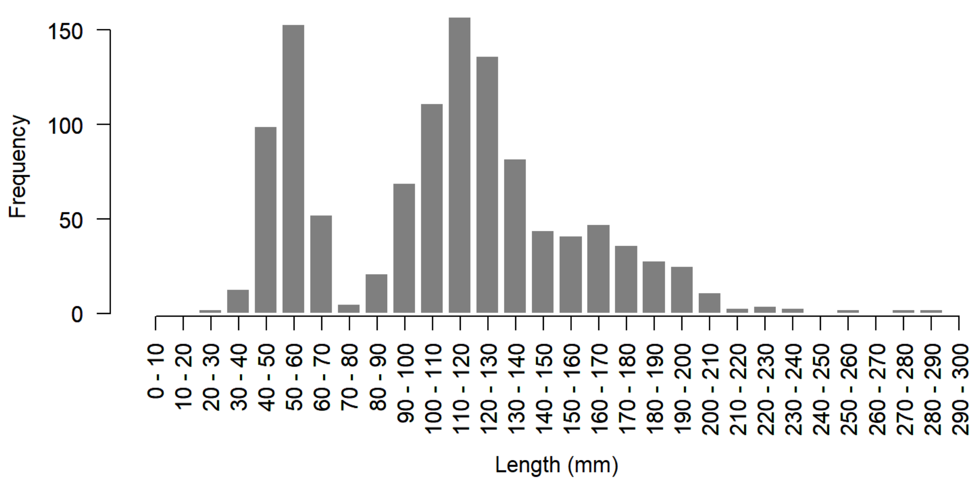


Figure 4. Length frequency histogram of all Brook Trout removed from Tyee Springs (N = 1145) in 2020 from August 12 to September 03.

2021

Brook Trout removal efforts in 2021 occurred over 14 days during July and August. A total of 2,314 Brook Trout were removed during this sampling period. The majority of Brook Trout (88%) were captured with the electrofishing tow-barge, which was the most effective method in all habitat units (Table 2). A backpack electrofisher was used in Units #2 and #3 and gillnets were deployed in Unit #3 only. The first two days of sampling consisted of scouting, equipment testing, planning, and gillnet experimentation. Our gillnet with the smallest mesh size (3/8 in. square) proved to be ineffective because its mesh was too large to effectively trap age-0 fish, yet small enough to accumulate debris and aquatic vegetation. Thus, after July 01 2021, only two gill nets were deployed (½ and ¾ inch mesh size) for the remainder of the field season.

Table 2. Number of Brook Trout removed by date, habitat unit and capture method in 2021. TB = tow-barge; EF = backpack electrofisher; GN = gillnets.

	Unit 1				Unit 2				Unit 3				
Date	**TB**	**EF**	**GN**	**Sum**	**TB**	**EF**	**GN**	**Sum**	**TB**	**EF**	**GN**	**Sum**	**Totals**
06/30/21	0	0	0	0	0	0	0	0	0	8	0	8	**8**
07/01/21	0	0	0	0	0	0	0	0	0	0	19	19	**19**
07/06/21	121	0	0	121	0	0	0	0	132	0	0	132	**253**
07/07/21	0	0	0	0	0	68	0	68	175	0	25	200	**268**
07/13/21	0	0	0	0	75	0	0	75	216	0	0	216	**291**
07/14/21	0	0	0	0	56	0	0	56	0	0	17	17	**73**
07/20/21	0	0	0	0	0	22	0	22	129	90	0	219	**241**
07/21/21	0	0	0	0	17	0	0	17	50	0	13	63	**80**
07/27/21	0	0	0	0	148	0	0	148	0	0	0	0	**148**
07/28/21	0	0	0	0	0	0	0	0	101	0	19	120	**120**
08/03/21	0	0	0	0	127	0	0	127	142	0	0	142	**269**
08/04/21	0	0	0	0	114	0	0	114	127	0	1	128	**242**
08/18/21	0	0	0	0	67	0	0	67	143	0	0	143	**210**
08/24/21	92	0	0	92	0	0	0	0	0	0	0	0	**92**
Totals	213	0	0	**213**	604	90	0	**694**	1215	98	94	**1407**	**2314**

Cells with zeros indicate no effort for that particular day, unit and sampling method.

The median fork length of captured Brook Trout was 112 mm, ranging from 22 to 228 mm (Figure 5; Figure 6). Fish lengths decreased on average as we sampled higher in the system, although Unit #3 yielded the widest range of fish lengths. Field personnel determined the sex of 388 randomly selected Brook Trout during the season, ranging in size from 95 to 288 mm. Of the total, 59% were female and 49% were male. Within Tyee Springs, Brook Trout >150 mm are more likely to be female which can bias sex ratio estimates for age-0 and age-1 fish. Thus, omitting all fish >150 mm from the analysis, the estimated sex ratio was 51% female (98 fish) and 49% male (96 fish) in 2021 (Figure 7).

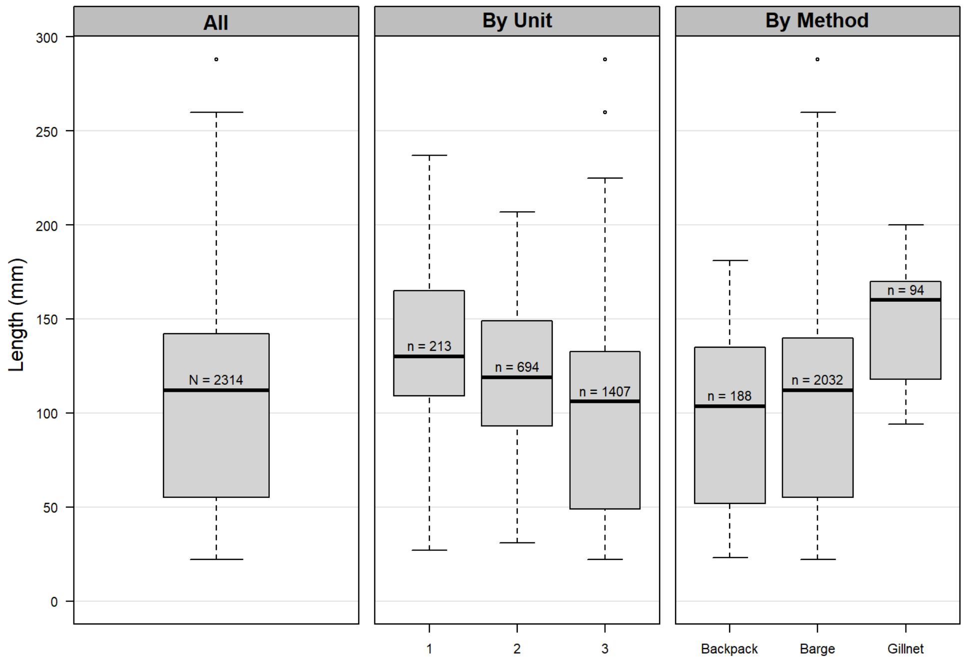


Figure 5. Boxplots of Brook Trout fork lengths from Tyee Springs removal efforts, all habitat and capture methods combined (left plot), by habitat unit (middle plot) and by capture method (right plot) in 2021. Boxes indicate the interquartile ranges (25% – 75%), whiskers indicate the total range (sans outliers), and circles indicate outliers (observations greater than 1.5 times the interquartile range).

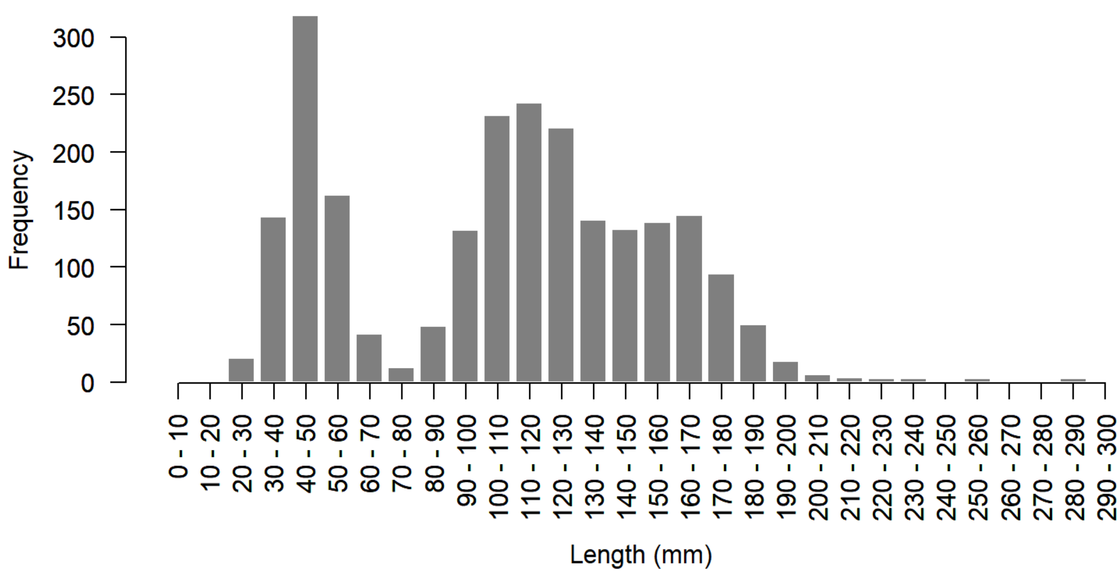


Figure 6. Length Frequency Histogram of all Brook Trout removed from Tyee Springs (N = 2,314) in 2021 from June 30 to August 24.

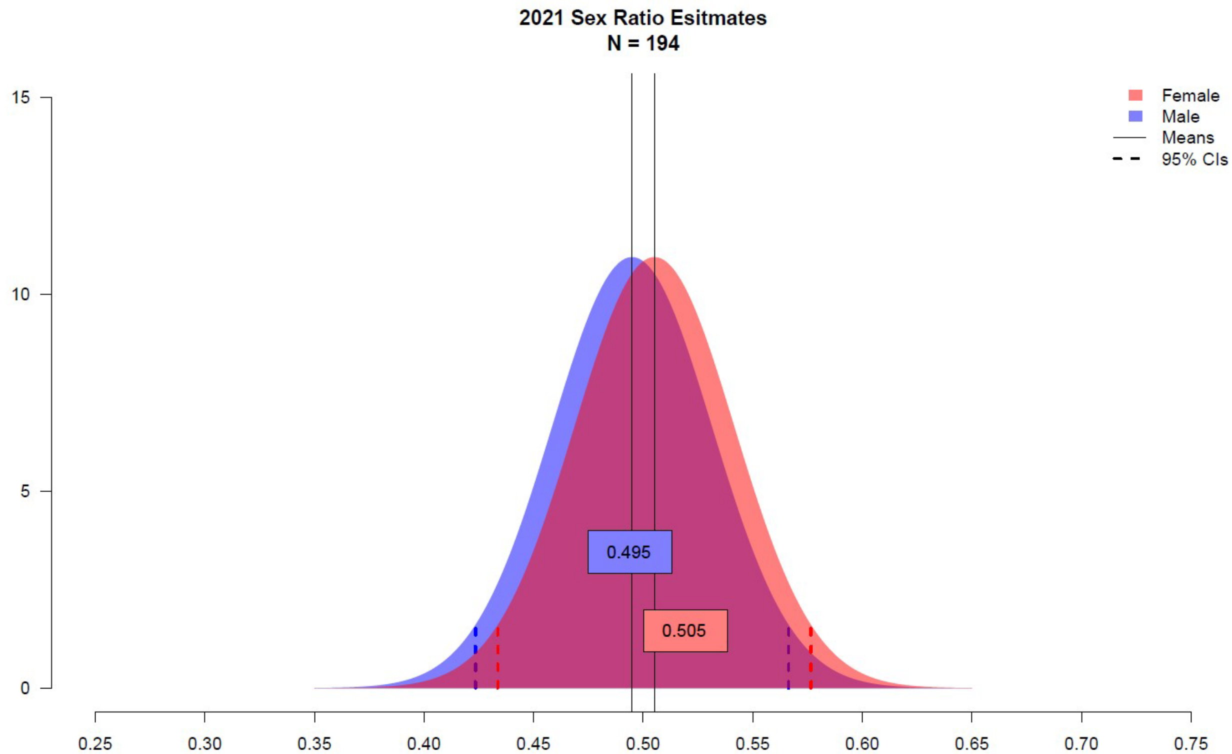


Figure 7. Sex ratio estimates from 194 Brook Trout (<150mm) captured in Tyee Springs, 2021.

M_{yy} Stocking

2020

A total of 600 M_{yy} Brook Trout were stocked in Tyee Springs at a rate of 200 fish per day, October 5 to October 7 2020. All 600 individuals were both adipose clipped and PIT tagged and average length in millimeters was 323.2 ± 21.9 (mean ± SD) at the time of tagging (Figure 8). Four hundred M_{yy} were stocked in the headwaters of Tyee Springs (Unit #3) and 200 fish were stocked in the deep water channel nearest Carson NFH (Unit #1). No M_{yy} fish were stocked into Unit #2 due to inaccessibility (Table 1).

2021

A total of 1,923 M_{yy} Brook Trout were stocked into the headwaters of Tyee Springs (Unit #3) over three days. All fish were adipose clipped and 999 were PIT tagged. Average length in millimeters of M_{yy} Brook Trout at the time of PIT tagging approximately one month prior to stocking was 163.7 ± 26.8 (mean ± SD) (Figure 8). All non-tagged fish (924) were released on the first stocking day (October 1 2021), 566 PIT tagged fish were released on the second day

(October 4 2021) and the remaining 433 PIT tagged fish were released on the third day (October 5 2021; Table 1).

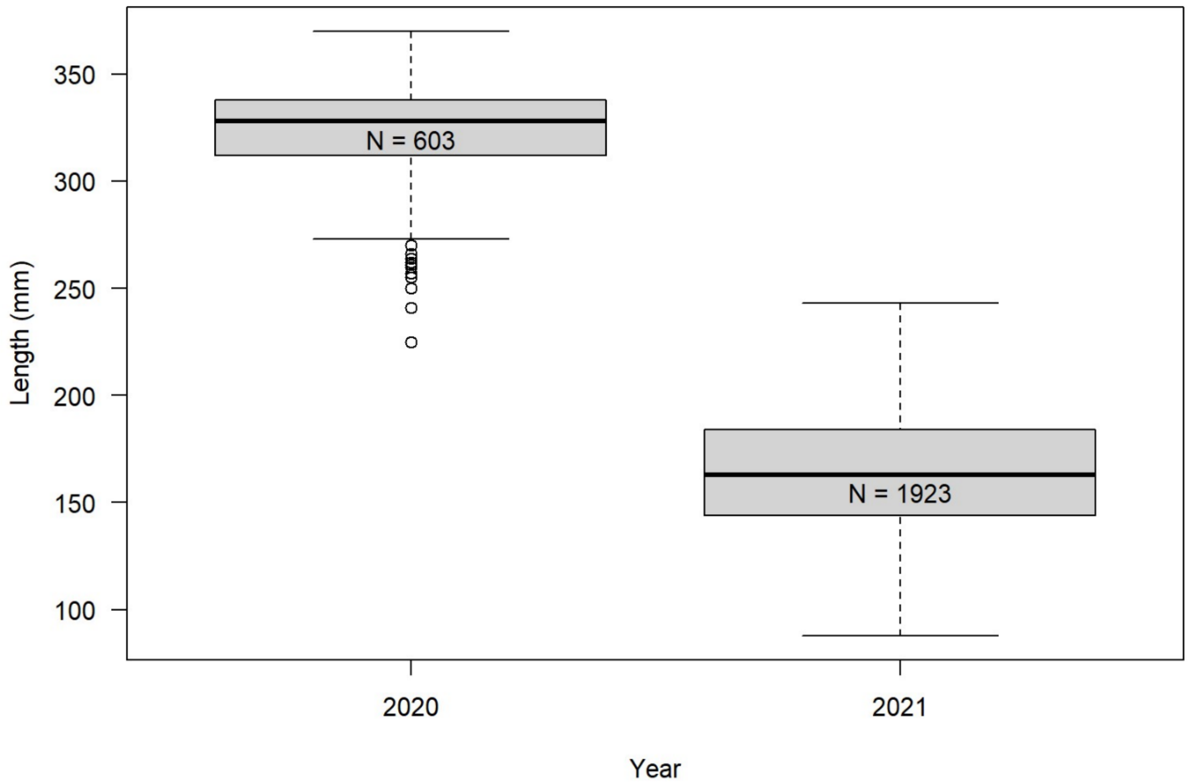


Figure 8. Boxplot of lengths of YY-males stocked in Tyee Springs in 2020 and 2021. Boxes indicate the interquartile ranges (25% – 75%), whiskers indicate the total range and circles indicate outliers (observations greater than 1.5 times the interquartile range).

Table 3. YY-Male Brook Trout stocking information for years 2020 and 2021. All M_{yy} were adipose fin clipped.

Date Stocked	M_{yy} Brood Year	Pit Tagged?	Stocking Location	Total M_{yy} Stocked
10/05/2020	2018	Yes	Unit #3	200
10/06/2020	2018	Yes	Unit #3	200
10/07/2020	2018	Yes	Unit #1	200
10/01/2021	2020	No	Unit #3	924
10/04/2021	2020	Yes	Unit #3	566
10/05/2021	2020	Yes	Unit #3	433

Monitoring

Post-stocking mortalities

In 2020, Carson NFH staff collected a total of 73 M_{yy} Brook Trout mortalities (12.1%). All mortalities were recovered within two days of stocking (October 7-8) at the hatchery intake grate (Unit #1). Fork lengths of mortalities were representative of the entire stocked population.

In 2021, hatchery staff recovered approximately 100 M_{yy} mortalities (~5.2%) atop the hatchery intake screen the day after the first stocking event (October 2). None of the recovered Brook Trout were PIT tagged and no other M_{yy} fish were recovered afterwards. All 2021 mortalities were sent to AFTC for examination.

Additional post-stocking mortality likely occurred in 2020 and 2021, but due to predation, scavenging, location and/or timing, the fish were never recovered and the total number of mortalities is unknown.

Post-stocking PIT detections

<u>2020</u>

We experimented with floating a PIT antenna from the headwaters downstream to the hatchery intake channel on November 04, 2020 and December 08, 2020. We detected 39 unique tags from a mark-recapture study conducted in 2018 and 2019 (Poirier et al. 2020) and 9 unique tags from M_{yy} Brook Trout stocked in early October 2020 (1.5%). Additionally, when we pulled the antenna out of the water at the intake channel (Unit #1), we detected six shed PIT tags laying in the grass. These tags were from five hatchery Chinook tagged at Carson NFH and one wild steelhead tagged in the Wind River (Table 4). River otters and scat are commonly seen in the surrounding area near the intake channel, so it is speculated that predation was the cause of these shed tags.

Table 4. Unique detections from floating PIT antenna in 2020.

Month/Day	Mark-Recap	M_{yy} 2020	Other
11/04	28	7	3
12/08	6	2	3
Total	**34**	**9**	**6**

<u>2021</u>

We deployed the floating PIT antenna a total of four days following the 2021 M_{yy} stocking event. Of the 999 PIT tagged individuals stocked in 2021, we detected 84 unique tags (8.4%). We also detected 6 tags from the stocking event that occurred in 2020, 108 tags from the 2018/19 mark-recapture study, and 23 miscellaneous tags consisting of 18 Carson NFH Chinook, 4 steelhead tagged in the Wind River, and 1 Spring Creek Hatchery Chinook (Table 5). Similar to 2020, tags originating from the Carson hatchery and Wind River were likely the result of predation. It is highly doubtful juvenile Chinook could make their way into Tyee Springs from hatchery raceways and we have yet to capture a Chinook or steelhead while conducting Brook Trout removals in Tyee Springs.

Table 5. Unique detections from floating PIT antenna in 2021.

Month/Day	Mark-Recap	M_{yy} 2020	M_{yy} 2021	Other
10/27	42	4	23	10
11/03	20	1	27	2
11/10	27	0	26	5
11/17	19	1	8	6
Total	**108**	**6**	**84**	**23**

Bypass Channel

2020 - 2021

The Carson NFH bypass channel opened on October 16, 2020 and closed on April 28, 2021. A total of three unique tags were detected during this operational period (Figure 9). All three tags (0.5 % of total stocked) were from M_{yy} Brook Trout released in 2020. It is possible more tags passed over the antennas during the detection period. One week after the channel opened on October 23, 2020, a tagged M_{yy} Brook Trout was discovered wedged underneath the first PIT antenna, resulting in a constant stream of repetitive information being logged into the transceiver. Due to non-ideal transceiver settings (unique-delay mode was turned off), data recorded during the first detection week in 2020 were overwritten and thereby lost.

2021 - 2022

In 2021 the bypass Channel opened on October 22 and closed on May 04, 2022. A total of 50 PIT tags were recorded over the detection period with the first detection occurring the day the channel opened and the last occurring on January 19, 2022 (Figure 9). All 50 of the detected tags (2.6%) were from the 2021 M_{yy} release. All fish lengths associated with detected PIT tags were representative of the stocked population.

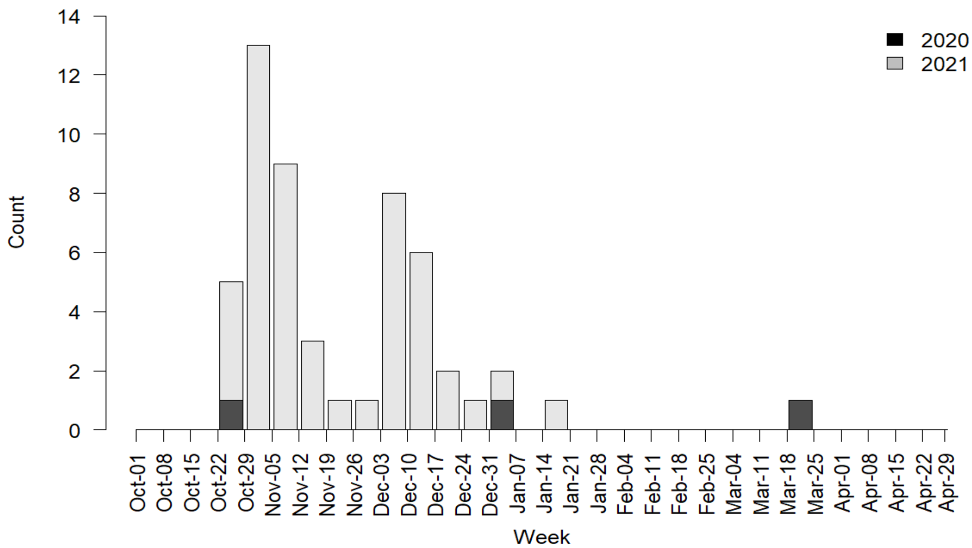


Figure 9. Weekly PIT detections in the Carson NFH bypass channel for the years 2020 (n = 3) and 2021 (n = 50).

Discussion

We encountered a number of unforeseen challenges during the first two years of the Tyee Springs M_{yy} project. Concerns about COVID-19 and measures to protect staff from potential exposure, limited staff assistance and shortened the field season in 2020. Permitting issues related to the unscreened bypass channel substantially reduced the total number of fish that could be stocked in Tyee Springs. Further, we observed unexpectedly high levels of post-stocking mortality potentially related to water chemistry, stocking location, predation or the age of M_{yy} at stocking. While these difficulties undoubtedly delayed the initial progress of the project, they also resulted in the overall improvement of our fish removal and stocking strategies.

The COVID-19 pandemic significantly delayed fish suppression efforts in 2020, resulting in much lower Brook Trout removal rates than expected. Additionally, we were unable to stock the desired number of M_{yy} Brook Trout in Tyee Springs (i.e., 3,000 fish) due to concerns with the unscreened bypass channel and potential for M_{yy} to escape Tyee Springs and enter the Wind River. We requested an informal Section 7 consultation with NOAA's National Marine Fisheries Service on the release of M_{yy} Brook Trout into Tyee Springs and provided NMFS with a Biological Evaluation of the project's effects. National Marine Fisheries Service found that stocking M_{yy} was not likely to adversely affect species or habitat listed under the Endangered Species Act and we received approval to stock a total of 600 M_{yy} Brook Trout in 2020 and 2,000 M_{yy} in 2021. We also observed a relatively high level of post-stocking mortality of M_{yy} Brook Trout in 2020 (≈12.1%), possibly due to differences in water chemistry between Abernathy FTC rearing water and Tyee Springs. Although M_{yy} Brook Trout were transported from Abernathy FTC in water obtained from Tyee springs, there was no prolonged acclimation period before stocking, other than during the three hour transport period. The difference in water hardness between Abernathy FTC (C_aCO_3 = 117 mg/L measured on 10/2019) and Tyee Sprigs (C_aCO_3 = 14 mg/L measured on 6/2018), may have contributed to high post-stocking mortality rates (Trushenski et al. 2019). In 2021, Abernathy FTC used a variety of acclimation strategies, including water mixing and adding salt to the transport water to minimize the stress associated with fish transport and differences in water chemistry. Post-stocking mortality was still somewhat high in 2021 (≈5.2%), but lower than levels observed in 2020. In future years, we plan to implement a slower stepwise water mixing acclimation strategy to further reduce post-stocking mortality. Mortality of M_{yy} Brook Trout was also potentially related to stocking location or predation. In 2020, 200 M_{yy} Brook Trout were stocked in Unit #1 immediately adjacent to the hatchery intake screen. All M_{yy} mortalities recovered from the intake screen (n=73) were from this stocking group. Hatchery staff suspect M_{yy} Brook Trout may have been attracted to the darkness of the intake screen outbuilding and were inadvertently caught on the screen. Additionally, Unit #1 is where hatchery staff regularly observe river otters and where we have detected M_{yy} PIT tags on the streambank near otter scat. In 2021, all M_{yy} fish were stocked in the headwaters of Tyee Springs (Unit #3) and hatchery staff turned the light on in the intake screen outbuilding the first few days after stocking to discourage fish from approaching the screen. Lastly, M_{yy} stocked in 2020 were age-1 (BY2018) fish versus age-0 (BY2020) fish in 2021. The age-1 M_{yy} were significantly larger than wild Brook Trout in Tyee Springs which could provide a competitive advantage over resident males in Tyee Springs (Armstrong and Caldwell 2021; Kennedy et al. 2018). However, during post-stocking monitoring with the floating PIT antenna, we observed age-1 M_{yy} fish grouped together in a large school rather than dispersed throughout Tyee Springs like age-0 M_{yy} fish. It is unclear whether this schooling behavior could have increased susceptibility to predation or impacted spawning with resident Brook Trout in Tyee Springs. No M_{yy} Brook Trout stocked in 2020 were recovered during fish removal efforts in 2021, indicating minimal if any overwinter survival. The majority of age-1 M_{yy} fish were presumably mature and ready to spawn a few months after stocking, but age specific survival is generally lower for age-1+ Brook Trout in Tyee Springs (see Poirier et al. 2020). In contrast, five age-0 M_{yy} Brook Trout stocked in 2021 were recovered during fish removal efforts in 2022, suggesting a low level of overwinter survival. Moving forward, we will continue to stock age-0 M_{yy} fish in Tyee Springs. They are less expensive to rear, easier to transport (i.e., one transport trip versus three), they may be less prone to predation immediately after stocking and there is evidence of higher overwinter survival, increasing the number of potential M_{yy} spawners the following year.

Despite challenges, the first two years implementing M_{yy} stocking into Tyee Springs were valuable in terms of learning and experimenting with different capture methodologies and monitoring strategies. Previous modeling work has shown that combining M_{yy} releases with annual fish suppression can reduce the overall time to eradication (Schill et al. 2017; Day et al. 2020; Poirier et al. 2020; Day et al. 2021). Thus, the majority of time and effort on this project has been allocated to Brook Trout suppression and refining capture techniques to maximize our effectiveness and efficiency. Based on removal totals, the electrofishing tow-barge has been the most effective sampling method in all three habitat units, capturing 74% of total Brook Trout in 2020 and 88% in 2021 (Table 1; Table 2). In 2021 we discovered that it was feasible and highly effective to operate the tow-barge in Unit #2 which likely accounts for the 14 percentage point increase from the year prior. Further, the tow-barge is able to fish shallow and deep water habitats equally well and we can run two anodes simultaneously, stunning more fish over a broader area. Backpack electrofishing did not capture as many fish relative to the tow-barge (25% in 2020 and 8% in 2021), but it was an indispensable tool for accessing areas the tow-barge could not reach such as small side channels, narrow inlets and shallow pools. In 2021, two gillnets (½ and ¾ inch mesh size) were deployed once per week in Unit #3. Although gillnets captured the least amount of Brook Trout compared to other methods (4% in 2021; Table 2), they were the most time-efficient capture method as they fished passively overnight and were relatively easy to deploy and retrieve. Minnow traps proved to be the least effective method for capturing Brook Trout in Tyee Springs and the method was quickly abandoned after the first few days of sampling in 2020. We also explored using a floating PIT antenna to monitor the movement of M_{yy} in the months after stocking. The survey was conducted in fall (late October to early December) to learn more about Brook Trout spawning, but we were unable to determine when and where spawning may be occurring based on our limited number of surveys. The read range of the antenna was approximately 0.5 m, so it was not as effective in Units #1 and #3 where water depths exceed 1.5 m. Nonetheless, the antenna was still a useful tool for locating shed tags, identifying dispersion behavior, weekly movement and annual survival of individual M_{yy} fish.

A PIT array was installed in the hatchery bypass channel to monitor the timing and abundance of PIT tagged Brook Trout potentially leaving Tyee Springs. The array detected a total of three M_{yy} Brook Trout in 2020 and 50 M_{yy} Brook Trout in 2021. Most of the fish that left Tyee Springs did so during the first two months the bypass channel was open (Figure 9). This time period also coincides with the heaviest amounts of precipitation and bypass channel flow. This emigration is concerning given the potential for M_{yy} Brook Trout to enter the Wind River where they could pose a threat to native fish populations. The loss of M_{yy} fish could also extend our eradication timeline. To address the issues associated with M_{yy} Brook Trout emigration to the Wind River, Carson NFH is working with the Washington Department of Fish and Wildlife (Yakima screen shop) to design and fabricate an inclined belt screen that will be installed in the bypass channel downstream from the hatchery intake screen building. The belt screen will be constructed to fill the entire width of the channel, blocking passage of Brook Trout that may be flushed into the bypass channel when it is open. Funding for the bypass screen was approved in June 2021 and installation is expected to occur in summer 2022. After the screening system is installed and Tyee Springs is a completely closed system, we expect to see even higher rates of M_{yy} introgression than those observed in other M_{yy} field evaluations; potentially reducing our time to eradication.

We conducted a population modeling exercise to evaluate the feasibility of stocking M_{yy} Brook Trout to eradicate resident Brook Trout in Tyee Springs (Poirier et al. 2020). Using population demographic information collected from the resident Brook Trout population (i.e., population abundance, age specific abundance, age specific survival, fecundity), model simulations estimated eradication of the resident population would occur after nine years on average if we suppressed 50% of the resident population annually, stocked 3,000 M_{yy} annually and survival of M_{yy} from release to spawning was 18% (Schill et al. 2017). Based on the estimated abundance of resident Brook Trout in Tyee Springs (7,140 fish in fall 2019), we hoped to remove approximately 3,500 fish prior to stocking to meet our goal. Due to unforeseen circumstances, our 2020 field season was abbreviated and we fell short of our 50% removal goal, only capturing 1,145 fish (~15%). We were also unable to meet our target 50% stocking goal due to concerns about the unscreened bypass channel. After updating the simulation model with 2020 stocking and suppression totals, the expected abundance of resident Brook Trout was 5,022 fish. In 2021, we successfully removed a total of 2,314 Brook Trout over the course of 14 days, giving us an estimated suppression rate of 46%. Based on the suppression and stocking rates that occurred in 2020 and 2021, as well as revising our original planned stocking rates to 2,000 M_{yy} annually, the model in its current state estimates eradication to occur after 14 years of stocking on average, i.e., the year 2033. It will be necessary to recalibrate the simulation model annually to incorporate any new information or unanticipated changes that may influence the eradication timeline.

Going forward, it will be important to evaluate the effectiveness of this project to refine the population model and adaptively manage our implementation strategies over time. For example, the model currently assumes that 18% of M_{yy} Brook Trout survive to the spawning period and have an equal probability of spawning with a female as resident males do. This 18% rate was taken from the literature (Schill et al. 2017) and may or may not be applicable to the Tyee Springs population. To assess M_{yy} spawning contribution in Tyee Springs, we randomly collected tissue samples from 100 M_{yy} Brook Trout, 100 adult resident Brook Trout and 400 young-of-year (i.e., age-0) Brook Trout during fish removal efforts in 2021. Tissue samples from adult fish are needed to characterize the genetic variation between M_{yy} and Tyee Springs Brook Trout populations, while samples from juvenile fish are used to determine whether they are M_{yy} progeny. The presence and proportion of M_{yy} progeny detected within our samples, will confirm the survival and spawning success of M_{yy} Brook Trout. Not only will this help inform the model, but it may also help guide future stocking strategies (e.g., number to stock, size/age-class to stock). Until we secure funding to analyze genetic samples we intend to determine the sex from a large subsample of Brook Trout captured during annual fish removals to track sex ratios over time. In 2021, males ranging in size from 100 to 150 millimeters (i.e., age-1) made up approximately 49% of the 194 Brook Trout we sexed. When we included Brook Trout over 150 mm, the sex ratio skewed toward more females (41% male, n = 388), suggesting a difference in either survival or growth between the males and females. In 2022 we should expect to see a shift in the sex ratio of age-1 fish if any of the 600 M_{yy} Brook Trout stocked in 2020 spawned successfully. These future field observations in tandem with genetic parentage analyses will provide key information to help calibrate the population model and allow us to measure the progress and potential success of M_{yy} stocking efforts in Tyee Springs.

Brook Trout population demographic parameters may be higher or lower than those estimated in 2019, which is why long term monitoring of the population will be an essential component of the project. We will continue to implement an adaptive management approach, using annual fish removal efforts to gather information about the M_{yy} and resident Brook Trout population to improve the model and attain a more accurate estimate of if/when complete eradication of Brook Trout is likely to occur in Tyee Springs. As the project progresses, we would like to address the following data gaps and implementation needs:

- Estimate the growth and overwinter survival of M_{yy} Brook Trout using fish re-captured during fish removal efforts.
- Investigate resident female fecundity and length at maturity.
- Assess the efficiency of population suppression techniques and how efficiency is affected by the gradual decline in abundance.

- Assess fish removal capture rates by age class.
- Expand post-stocking monitoring to better understand brook trout spawn timing and locations in Tyee Springs.
- Revise the population abundance estimate in Tyee Springs.
- Test the effectiveness of slower, stepwise water mixing acclimation strategy to reduce M_{yy} post-stocking mortality.
- Process genetic samples to assess M_{yy} reproductive success.

The use of M_{yy} Brook Trout as a biological control strategy remains relatively new. Evaluation of the techniques effectiveness (or potential shortcomings) is important to inform the science and refine field implementation methods for both current and future applications. If the eradication of Brook Trout is successful in Tyee Springs, the Service will save money by eliminating the cost of maintaining Brook Trout exclusion screens at Carson NHF. Successful eradication will also remove the threat of inadvertently releasing Brook Trout into the Wind River and will eliminate a vector for BKD transmission to Chinook salmon in Carson NFH. This proof-of-concept evaluation can also help the Service and its partners determine if and how the M_{yy} technique can be used to control nuisance Brook Trout populations in other locations and set the stage for evaluating whether the approach may be useful with other invasive fish species, like common carp, which have proven very difficult to control in places like Malheur National Wildlife Refuge.

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Appendix

Tyee Springs habitat Unit #1: hatchery intake channel (left), habitat Unit #2: free flowing stream (middle) and habitat Unit #3: headwaters of Tyee Springs (right).

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