## U.S. Fish and Wildlife Service <br> Columbia River Fish and Wildlife Conservation Office

# Feasibility Assessment of Stocking YY Males to Eradicate Nonnative Brook Trout from Tyee Springs 

FY 2019 Annual Report


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On the cover: Brook Trout captured in Tyee Springs at Carson National Fish Hatchery in Carson, Washington. Photo by Patrick Cooney.

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# Feasibility Assessment of Stocking YY Males to Eradicate Nonnative Brook Trout from Tyee Springs 

2019 ANNUAL REPORT<br>Jennifer Poirier, Brian Davis, and Julianne Harris<br>U.S. Fish and Wildlife Service<br>Columbia River Fish and Wildlife Conservation Office<br>1211 SE Cardinal Court, Suite 100<br>Vancouver, WA 98683

## Abstract

An established population of nonnative Brook Trout resides in Tyee Springs directly upstream from Carson National Fish Hatchery. The potential for Brook Trout to escape from Tyee Springs and enter the hatchery is a concern because the hatchery releases spring Chinook yearlings into the Wind River as well as the South Fork Walla Walla River, a regional stronghold for ESAlisted Bull Trout. Past attempts to suppress the Brook Trout population have proven unsuccessful at eradicating the unwanted population. The Trojan Y Chromosome technique is a biological control strategy that is gaining popularity as an invasive fish management tool. This method involves producing male Brook Trout with two Y-chromosomes ( $\mathrm{M}_{\mathrm{yy}}$ ) which are then released into the population targeted for eradication. Offspring of YY males and resident females (XX) are all male (XY), so eventually the population becomes skewed toward a single sex, leading to extirpation of the target population. The Columbia River Fish and Wildlife Conservation Office, Abernathy Fish Technology Center, and Carson National Fish Hatchery initiated a collaborative proof-of-concept assessment of stocking $\mathrm{M}_{\mathrm{yy}}$ Brook Trout to eradicate nonnative Brook Trout in Tyee Springs. The purpose of this study was to collect demographic information from the resident Brook Trout population and perform a population simulation exercise to evaluate the feasibility of using $\mathrm{M}_{\mathrm{yy}}$ fish to eradicate nonnative Brook Trout in Tyee Springs. Results from simulations indicate that eradication of nonnative Brook Trout is possible using the Trojan Ychromosome technique and that time to eradication generally decreases with increases in annual fish suppression rates. Consistent with other $\mathrm{M}_{\mathrm{yy}}$ simulation work, stocking Myy fish at $50 \%$ of the resident Brook Trout population ( 3,000 fish) and suppressing $50 \%$ of the resident population annually would lead to an $80 \%$ probability of eradication within nine years. Given suppression rates will likely decline over time, we tested suppression rate as a function of abundance, incrementally reducing the suppression rate as resident Brook Trout abundance declines. Under these conditions, the estimated number of years to eradication increased to 13 years versus 9 years when suppression was held at a constant $50 \%$. A possible alternative to expending more effort to suppress the resident population would be to increase $\mathrm{M}_{\mathrm{yy}}$ stocking rates. However, sensitivity analyses indicate that there is no difference in time to eradication between stocking

3,000 or $7,000 \mathrm{M}_{\mathrm{yy}}$ Brook Trout at suppression rates $\geq 25 \%$. Given uncertainty surrounding the effectiveness of fish removal efforts along with survival and reproductive fitness of $\mathrm{M}_{\mathrm{yy}}$ fish once they are stocked into Tyee Springs, it will be necessary to utilize an adaptive management approach for the duration of the project. We recommend using annual fish removal events as an opportunity to monitor the population and collect additional information to update and improve the population model that will in turn inform and guide future fish removal and stocking efforts. Successful eradication of nonnative Brook Trout in Tyee Springs will remove a direct threat to Carson National Fish Hatchery and ESA-listed bull trout in the South Fork Walla Walla River. This project will help inform invasive fish eradication efforts and will be one of the first studies to assess how well the simulation model predicts the time to eradication of a nonnative Brook Trout population in a natural stream using the Trojan Y Chromosome technique.

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## Introduction

Carson National Fish Hatchery (NFH) in south-central Washington annually rears about 1.4 million spring Chinook salmon, of which 250,000 are released into the Walla Walla River system which is a regional stronghold for ESA-listed bull trout. The water source for Carson NFH, Tyee Springs, contains a population of introduced Brook Trout. This raises the possibility that Brook Trout could enter the hatchery and inadvertently be transferred with juvenile Chinook salmon into streams containing bull trout. Nonnative Brook Trout are considered a threat to bull trout where the two co-occur because of hybridization, competition, and potential predation (Rieman et al. 2006; Dunham et al. 2002). Carson NFH currently uses a self-cleaning screen system with 1.5 mm profile bars to keep Brook Trout from entering hatchery raceways, but this structure requires frequent maintenance and Brook Trout have been observed in hatchery ponds as recently as 2013. Consequently, in some years hatchery and other U.S. Fish and Wildlife Service staff have had to physically sort through fish at the time of release to ensure Brook Trout were not accidentally transferred with the spring Chinook salmon. Carson NFH also operates a secondary bypass channel that diverts water from lower Tyee Springs (i.e. hatchery intake) to the Wind River during late fall through early spring to prevent flooding. Brook Trout may be flushed into the Wind River during this period, posing a potential threat to native fish populations. Lastly, Brook Trout in Tyee Springs carry low levels of Bacterial Kidney Disease (BKD) and may transmit the disease to Chinook salmon in Carson NFH or native salmonids in the Wind River.

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 ample refuge for fish to escape capture (USFWS 2004).

An alternative biological control strategy that could eradicate Brook Trout in Tyee Springs is the so-called 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 $\mathrm{M}_{\mathrm{yy}}$ ) and resident females (XX) are all male (XY), so eventually the population becomes skewed toward a single sex, theoretically leading to extirpation of the target population due to reproductive failure. The Idaho Department
of Fish and Game has developed a broodstock of $\mathrm{M}_{\mathrm{yy}}$ Brook Trout for experimental use and the first field stocking trials are currently underway with encouraging preliminary results (see Kennedy et al. 2018).

Despite the promise the $\mathrm{M}_{\mathrm{yy}}$ strategy holds for nonnative fish eradication, there is a good deal of uncertainty regarding its likelihood of success and the potential time and effort needed to achieve total eradication of the undesirable population. Field trials have not progressed to the point where the effectiveness of the $\mathrm{M}_{\mathrm{yy}}$ technique can be assured. To reduce this uncertainty and assess the potential effectiveness of the technique, we employed a population simulation model (described in Schill et al. 2017) to evaluate the efficacy of the $\mathrm{M}_{\mathrm{yy}}$ technique as a potential eradication strategy for nonnative Brook Trout in Tyee Springs. The objectives of our study were to determine the population structure and abundance of the resident Brook Trout population currently inhabiting Tyee Springs and use Brook Trout life history parameters to populate a stochastic population matrix model. This model was used to assess the feasibility of Brook Trout eradication by stocking $\mathrm{M}_{\mathrm{yy}}$ in Tyee Springs in terms of how stocking rates and removal rates affect years to eradication of Brook Trout.

The Columbia River Fish and Wildlife Conservation Office (FWCO), Abernathy Fish Technology Center (FTC), and Carson National Fish Hatchery (NFH) initiated a collaborative proof-of-concept assessment of stocking $\mathrm{M}_{\mathrm{yy}}$ Brook Trout to eradicate nonnative Brook Trout in Tyee Springs. This report presents results of population sampling and simulation modeling conducted by FWCO personnel in 2018 and 2019.

## 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. Carson NFH, which began operation in 1937, was constructed to utilize Tyee Springs as its primary water source, providing year round flow at a constant $6.6^{\circ} \mathrm{C}$. 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 secondary 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, likely from previous stocking efforts. 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 Population Sampling

Mark-recapture methods were used to estimate population abundance, age structure, growth, and inter-annual survival of resident Brook Trout in Tyee Springs. Sampling was conducted over a period of two consecutive days in fall, 2018 (Nov. 14-15), spring, 2019 (May 21-22), and fall, 2019 (Oct. 16-17). The stream was divided into three sampling units (based on habitat characteristics) and fish were captured using an electrofishing tow barge or backpack electrofisher depending on the water depth of each unit.

The 48 m long hatchery intake channel on lower Tyee Springs (Unit 1; Figure 2), was sampled using a Smith-Root electrofishing tow barge. A total of four passes were made in the channel, each consisting of pulling the barge from bank to bank either upstream or downstream in a zigzag fashion. The middle 400 m 'stream-like' portion of Tyee Springs (Unit 2; Figure 3), was sampled using a Smith-Root backpack electrofisher (model LR-24 or APEX). Field personnel made a single pass, moving from downstream to upstream during each sample event. Upper Tyee Springs (Unit 3; Figure 4), is a spring fed lake that was sampled using a Smith-Root electrofishing tow barge. Field crews made a single pass from downstream to upstream pushing the barge along the bank margins and through wadeable areas.

All captured fish were anesthetized in a solution of tricaine methanesulfonate (MS-222), measured (fork length, mm), weighed (g), and scanned for a PIT tag or external mark. Untagged fish greater than $\approx 60 \mathrm{~mm}$ in length were implanted with a 12 mm FDX PIT tag, while fish less than 60 mm in length were given an adipose fin clip. Pelvic fin clips were taken from a subsample of captured fish $(\mathrm{N}=400)$ and stored in ethanol for future genetic analysis. Following the collection of biological data, all fish were held in a floating net pen or aerated bucket and released into their respective sampling unit at the end of each day.

## PIT tag antenna array

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 in order to prevent flooding of the hatchery facility. The bypass channel is in use for approximately six months of the year and is not currently screened. The bypass channel is 'opened' by removing dam boards, allowing surface water from Tyee Springs to drop three feet into the channel. 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 $4-\mathrm{ft} \mathrm{x} 4$ - ft channel during peak flows (Figure 5). 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. Both PIT array installation and population sampling occurred prior to opening the bypass channel on October 21, 2019 and continued to operate until the bypass channel was closed on May 1, 2020. During operation, intake velocities were regulated as needed by removing or replacing dam boards at the head of the bypass channel.

## Demographic Rate Estimation

We estimated demographic rates to populate an stage-classified stochastic simulation model developed by Schill et al. (2017) to assess the effects of a range of stocking rates and suppression rates on the estimated time to eradication of invasive Brook Trout in Tyee Springs (see below). We estimated demographic rates by Bayesian methods using JAGS software (Plummer 2003) and package jagsUI (function autojags; Kellner 2018) called from Program R (R Core Team 2013). We sampled three chains with an adaption of 1,000 , a burn-in of 5,000 , and an iteration interval of 10,000 . We saved enough iterations to meet convergence, as assess by Rhat scores of less than 1.1 for all estimated and derived parameters (Gelman and Hill 2007; Kéry and Schaub 2012). For each estimated and derived parameter, we considered the median of the posterior distribution as the expected value and the $95 \%$ credible interval (" $95 \%$ ") as an assessment of variability. We assessed differences among parameter estimates when $95 \%$ credible intervals did not overlap and when $95 \%$ credible intervals on slopes did not overlap zero. Prior distributions were selected to be generally uninformative (specific priors used are reported below with the corresponding analysis).

Since we measured fork length, but did not assess age, for our PIT-tagged Brook Trout in Tyee Springs, we used a von Bertalanffy growth model to examine growth and estimate expected fork length-at-age. We needed expected fork-length-at-age to estimate abundance of individuals in each age class and expected survival rate for those age classes to populate the age-structured stochastic simulation model. The standard von Bertalanffy growth model estimates three parameters: $k$ (growth rate coefficient), $L_{\infty}$ (asymptotic length) and $t_{0}$ (hypothetical age when length is zero) to describe somatic growth of fish in a population (Quinn \& Deriso 1999; Haddon 2001). Fabens (1965) transformed the standard von Bertalanffy model which expresses mean length as a function of age to assess growth by change in length over time; thus, the Fabens (1965) model can be used with mark-recapture data as opposed to age data. We used data on fork length (in mm) of PIT-tagged Brook Trout captured on either two or three of the three sampling occasions ( $L_{i, j}$ ) to estimate von Bertalanffy growth parameters (i.e., $L_{\infty}$ and $k$ ) and to estimate expected length $\left(E_{i, j}\right)$ for each individual $(i)$ on each occasion $(j)$ :

$$
E_{i, j}=L_{\infty} *\left(1-\exp \left(-k *\left(A_{i}+t_{i, j}\right)\right)\right)
$$

Where $A_{i}$ is the estimated age at tagging minus $t_{0}$ for fish $i$, and $t_{i, j}$ is the measured period (in years) at large for fish $i$ between tagging and time $j$ and is estimated by a gamma distribution:

$$
A_{i} \sim \operatorname{gamma}\left(a_{1}, a_{2}\right)
$$

Where $a_{1}$ and $a_{2}$ were hyper-parameters estimated by uniform priors (over the range of 0-100). The prior for $L_{\infty}$ was a normal distribution (mean $=300$; variance $=10,000$ ) and the prior for $k$ was a uniform distribution (over the range of 0-2). Each measured length ( $L_{i, j}$ ) was from a normal distribution with mean expected length $E_{i, j}$ and an estimated standard deviation ( $\sigma$ ) representing model error, which includes both measurement error (i.e., error in measuring fish length) and process error (i.e., lack of model fit):

$$
L_{i, j} \sim \operatorname{Normal}\left(E_{i, j}, \sigma\right)
$$

The prior for $\sigma$ was also a uniform (over the range of $0-100$ ). As the age of marked individuals was unknown, we could not directly estimate $t_{0}$; thus, we estimated it by the following:

$$
t_{0}=\log \left(1-\left(\frac{12.8}{L_{\infty}}\right)\right) / k
$$

Where 12.8 mm is the assumed fork length of a Brook Trout larvae at hatch, as measured in the laboratory (Engle 2007). With estimates of $L_{\infty}, k$ and $t_{0}$, we could estimate expected length-atage $(t)$ in years by the following:

$$
E_{t}=L_{\infty} *\left(1-\exp \left(-k *\left(t-t_{0}\right)\right)\right)
$$

We estimated abundance, by fork length, of Brook Trout in Tyee Springs for the three sampling occasions by logistic regression. For each sampling occasion (i.e., November 2018 to May 2019 or May 2019 to October 2019), PIT-tagged Brook Trout that were tagged or captured on the first day of sampling were considered the marked portion of the population. Marked individuals that were recaptured on the second day of sampling $\left(R_{i}\right)$ were modeled using a Bernoulli distribution as a function of the probability of being recaptured $\left(r_{i}\right)$ :

$$
R_{i} \sim \text { Bernoulli }\left(r_{i}\right)
$$

We used the logit link to examine recapture probability as a function of the individual's true fork length $\left(L_{i, j}\right)$, as measured when captured on the first sampling day, and the collection site in Tyee Springs $\left(C_{i, j}\right)$ :

$$
\operatorname{logit}\left(r_{i}\right)=\gamma_{0}+\gamma_{1}\left[L_{i}\right]+\gamma_{2}\left[C_{i}\right]
$$

Where $\gamma_{0}$ is the intercept for recapture probability and $\gamma_{1}$ and $\gamma_{2}$ are slopes. We included collection site because we thought detection probability could potentially differ among sites based on differences in habitat both among sites and among sampling occasions (e.g., width, depth, water clarity, etc.), as well as differences in electrofishing sampling methods among sites (i.e., number of passes barge vs. backpack). We included one slope for the effect of fork length on recapture probability estimated using information from all sampling occasions but included independent slope estimates for collection site and intercept for the three sampling occasions. Within the model, we derived estimates of recapture probability by 10 mm fork length bin for each collection site. We then calculated expected abundance for each collection site for each 10 mm fork length bin by dividing the actual number of Brook Trout in that fork length bin captured on day two of the sampling occasion, by the recapture probability for that fork length bin and collection site. We summed estimates from all collection sites to obtain expected abundance, by 10 mm fork length bin, for each sampling occasion. In addition, we summed abundance estimates for all fork length bins to obtain total abundance estimates for each sampling occasion $(N)$, as well as estimated abundance by age class $\left(n_{t}\right)$, based on expected length-at-age (i.e., $E_{t}$ ) from growth modeling (described above).

We also estimated apparent survival ( $\phi$ ) and detection probability ( $p$ ) of PIT-tagged Brook Trout, by fork length, in Tyee Springs using a Cormack-Jolly-Seber (CJS) approach (Cormack 1964; Jolly 1965; Seber 1965). The CJS model is hierarchical and uses patterns of detections of tagged individuals over sampling occasions to estimate apparent survival over occasion intervals, as well as detection probability. The CJS model estimates "apparent survival" because the model cannot distinguish mortality from emigration. Before starting this study, we thought Tyee Springs was a closed system; however, we later discovered emigration was possible via the hatchery bypass channel, although immigration was not. We have limited information that a low level of emigration is occurring; thus, our CJS estimates of survival are biased low, although likely not substantially. In this study, we PIT-tagged Brook Trout on the three sampling occasions (November 2018, May 2019, and October 2018). On each sampling occasion, we tagged and potentially recaptured (except during the first sampling day) fish on two consecutive sampling days. We assumed no mortality or movement over the two sampling days. Thus, we produced two estimates of survival: the first from November 2018 to May 2019 (winter survival interval) and the second from May 2019 to October 2019 (summer survival interval). In this model, the probability that an individual Brook Trout $(i)$ was alive $\left(z_{i, j}\right)$ on a given sampling occasion $(j)$ after it was PIT-tagged was estimated by a Bernoulli distribution:
$z_{i, j} \sim$ Bernoulli $\left(\mu_{i, j}\right)$

Where $\mu_{i, j}$ is the product of the probability of being alive at the start of the previous sampling occasion (i.e., $z_{i, j-1}$ ) and the probability of surviving the previous interval (i.e., $\phi_{i, j-1}$ ). We used the logit link to examine survival for an individual over the previous sampling occasion $\left(\phi_{i, j-1}\right)$ as a function of the individual's true (if recaptured) or estimated (if not recaptured) fork length in the previous sampling occasion $\left(L_{i, j-1}\right)$ :

$$
\operatorname{logit}\left(\phi_{i, j-1}\right)=\alpha_{0, j-1}+\alpha_{1}\left[L_{i, j-1}\right]
$$

Where $\alpha_{0, j-1}$ is the intercept for survival over the interval from $j-1$ to $j$ (i.e., from November 2018 to May 2019 or May 2019 to October 2019) and $\alpha_{1}$ is the slope for the effect of fork length on survival. When recaptured during either day of a sampling occasion, tagged Brook Trout were measured for fork length ( mm ). If not recaptured, we estimate the expected fork length for an individual ( $i$ ) during the occasion $(j)$ using its fork length at the start of the previous interval and results from our growth modeling:

$$
L_{i, j}=\left(L_{\infty}-L_{i, j-1}\right) *(1-\exp (-k * t))+L_{i, j-1}
$$

Where $t$ is the time (in years) of the interval from occasion $j-1$ to occasion $j$.

Actual detections of each PIT-tagged Brook Trout ( $y_{i, j, d}$ ) on each sampling occasion $(j)$ and consecutive day $(d)$ were modeled using a Bernoulli distribution as a function of the probability of being recaptured $\left(P_{i, j, d}\right)$ :

$$
y_{i, j, d} \sim \operatorname{Bernoulli}\left(P_{i, j, d}\right)
$$

The probability an individual is recaptured ( $P_{i, j, d}$ ) on a given day $(d)$ during a subsequent sampling occasion is a function of the probability that the individual is alive $\left(z_{i, j}\right)$ and recapture probability on that sampling occasion and day ( $p_{i, j, d}$ ):

$$
P_{i, j, d}=z_{i, j} * p_{i, j, d}
$$

Based on abundance estimation results (i.e., slopes for effects of fork length and collection site), we used the logit link to examine detection probability as a function of the individual's true (if recaptured) or estimated (if not recaptured) fork length at the start of the sampling occasion $\left(L_{i, j}\right)$, as well as the collection site at the start of that interval $\left(C_{i, j}\right)$ :

$$
\operatorname{logit}\left(p_{i, j, d}\right)=\beta_{0}+\beta_{1}\left[L_{i, j}\right]+\beta_{2}\left[C_{i, j}\right]
$$

Where $\beta_{0}$ is the intercept for detection probability and $\beta_{1}$ and $\beta_{2}$ are slopes. We observed minimal movement among sites; thus, if not detected during a sampling occasion, we assumed an individual was still in the site where it was previously found. Within the model, we derived estimates of survival by 10 mm fork length bin for the winter $\left(\phi_{1}\right)$ and summer intervals $\left(\phi_{2}\right)$. We also calculated estimates of annual survival $\left(S_{f}\right)$ by 10 mm fork length bin $(f)$ by correcting the winter and summer estimates to account for the whole year:

$$
S_{f}=\left(\phi_{1, f} * \phi_{2, f}\right)^{1.09}
$$

We used information on expected length-at-age $\left(E_{t}\right)$ from our growth modeling to obtain expected survival for each age class, as needed to populate the age-structured stochastic simulation model to estimate expected time to eradication of Brook Trout in Tyee Springs.

## Model Simulations

Population demographic estimates (stage specific - survival rates, fertility rates, and abundance) were used to inform a stage-classified matrix simulation model (Figure 6). The model includes three stages representing Brook Trout of ages zero, one, and two or older. Projection intervals ( t , $t+1$ ) were defined as one year from each spawning period. Simulations were designed to estimate the number years to eradicate Tyee Springs Brook Trout and how different removal and $\mathrm{M}_{\mathrm{yy}}$ stocking strategies will affect eradication timing. Simulations were modeled as stochastic projections to predict how random variation in fecundity, survival rates, suppression rates, and $\mathrm{M}_{\mathrm{yy}}$ spawning success will affect projection outcomes.

Annual suppression rates (i.e., Brook Trout removal rates) were modeled as negative exponential curves to allow for testing changes in capture efficiency as abundance decreases. Suppression rate parameters were held at constant values during simulations and were varied during sensitivity analyses to test suppression rates ranging from 5 to 95 percent. Suppression rates were given an arbitrary standard deviation of 2.5 percent. As we suppress the Brook Trout population annually, new data will inform capture efficiency estimates and associated variance for future projection modeling.
$\mathrm{M}_{\mathrm{yy}}$ stocking scenarios were evaluated at rates ranging from 500 to 9500 individuals per year. Individual $\mathrm{M}_{\mathrm{y}}$ Brook Trout survival was drawn from random Bernoulli trials with the assumption that $\mathrm{M}_{\mathrm{yy}}$ Brook Trout in Tyee Springs will survive at a rate of $18 \%$ from stocking to their first spawning period (Kennedy et al. 2018). Preliminary data from concurrent studies have estimated annual survival of stocked $\mathrm{M}_{\mathrm{yy}}$ Brook Trout between $3 \%$ and $7.8 \%$ (YY Brook Trout Technical Team 2019). For the purposes of this model, annual survival of $\mathrm{M}_{\mathrm{yy}}$ individuals was assumed to be $5 \%$ from their first spawning period to their second and $1 \%$ from their second to third. Contribution of $\mathrm{M}_{\mathrm{yy}}$ Brook Trout to the spawning population was modeled as a
hypergeometric distribution to account for the number of $\mathrm{M}_{\mathrm{yy}}$ and naturally produced males competing for available females. $\mathrm{M}_{\mathrm{yy}}$ and naturally produced males were assumed to have an equal probability of spawning success. Spawning simulations between $\mathrm{M}_{\mathrm{yy}}$ and female individuals were coded to result in progeny consisting of 100 percent males, whereas spawning between two naturally produced fish resulted in random draws from a Bernoulli distribution with a $50 \%$ sex rate.

Fertility rate estimates $(F)$ were determined algebraically using November, 2018 abundance estimates of age-1 and age-2+ Brook Trout ( $N_{t, \text { age }>0}$ ); sex ratio ( $S R$ [assumed to be 0.5 ]); age- 0 survival estimates $\left(S_{\text {age }}\right)$; and 2019 abundance estimates of age- $0\left(N_{t+1, a g e 0}\right)$.

$$
F=\frac{N_{t+1, \text { age } 0}}{N_{t, \text { age }>0} * S R * S_{\text {age } 0}}
$$

Total fertility rate (number of age-0 fish produced per female) was then calculated and divided between age- 1 and age- $2+$ life stages disproportionately. The age- 1 fertility rate was set proportionally lower than age- $2+$ based on literature to account for lower fecundity and lower maturity rates (Kennedy et al. 2003; Vladykov, Legendre 1940). Total number of age-0 Brook Trout were modeled as a Poisson distribution where lambda is equal to the product of the total number of females and fertility rate $(F R)$ per adult stage.

$$
N_{t+1, \text { age } 0} \sim \operatorname{Possion}\left(F_{\text {age }} * N_{t, \text { age }}\right)
$$

## Sensitivity Analyses

The population model was evaluated with suppression rates fixed at $50 \%$ and $25 \%$ (per every age class), while stocking totals were held at 3,000 to compare how the two different fish removal efficiencies will affect estimated years until eradication occurs. These levels were selected based on analyses conducted by Schill et al. (2017) that found a $50 \%$ stocking rate and $50 \%$ suppression rate could lead to extirpation as low as 2-4 years. A one-way sensitivity analysis was conducted, varying one model parameter from reasonable low to high values while all other parameters were held at their mean value. The estimated number of years until eradication occurs was recorded for every parameter perturbation. A two-way sensitivity analysis was conducted between suppression rates and annual stocking rates. We chose these two parameters because they can be directly controlled. Suppression was varied from $5 \%$ to $95 \%$ while stocking rates were varied from 500 to 9500 individuals. These parameters were sequenced to a length of 10 , then every possible combination $(\mathrm{N}=100)$ of these two parameters were evaluated while all other parameters were held constant at their mean. Simulations ran for 1000 iterations to accurately capture variation due to demographic stochasticity. All modeling and analyses were conducted in R version 3.6.1 ( R Core Team 2020).

## Results

## Tyee Springs Population Sampling

We tagged 1,798 Brook Trout and recaptured 336 over the three mark/recapture sampling events (Table 1). The majority of fish were captured in Unit 3 (spring fed lake - barge electrofisher) and the highest capture efficiencies occurred in Unit 1 (hatchery intake channel - barge electrofisher). Unit 2 (stream type - backpack electrofisher) yielded the lowest total numbers of Brook Trout and the lowest capture efficiencies.

Brook Trout movement between designated habitat units was minimal. No movement was observed between consecutive mark/recapture days for each seasonal sampling event. We did not observe any movement between the first sampling season (fall 2018) and the second (spring 2019). All observable movement occurred between the second (spring 2019) and third (fall 2019) sampling events and only consisted of five individuals (three tagged in spring 2019 and two tagged in fall 2018). All five fish that moved between units were tagged in either Unit 1 or Unit 2; all other possible movement combinations occurred (1 to 2,2 to 1,1 to 3 and 2 to 3 ).

Table 1. PIT mark/recapture totals for three sampling periods (fall 2018, spring 2019, and fall 2019).

| Sampling Event | Tag/Recap | Unit 1 |  | Unit 2 |
| :---: | :--- | :--- | :--- | :--- |
| Unit 3 |  |  |  |  |
| Fall 2018 | Tagged | 187 | 129 | 337 |
|  | Recaptured | 63 | 3 | 21 |
| Spring 2019** | Tagged | 70 | 92 | 199 |
|  | Recaptured | 69 | 12 | 17 |
| Fall 2019** | Tagged | 145 | 182 | 457 |
|  | Recaptured | 92 | 37 | 55 |
| All | Tagged | 402 | 403 | 993 |
|  | Recaptured | 140 | 91 | 74 |

**Recaptures include tags from previous sampling events.

## PIT tag antenna array

The bypass channel PIT array detected a total of 24 individual PIT tags. Seventy-five percent of these tags were detected from January to March which coincides with months of heavier precipitation (Figure 7). Of the 24 detected tags, 8 originated from the fall 2018 tagging event, 5 from spring 2019 and 11 were from fall 2019. Seventeen of the tags ( $\sim 71 \%$ ) originated from Unit 1, which is the unit closest to the bypass channel. Every age-stage/size-class available for detection was represented emigrating to the Wind River including an individual that measured 77
millimeters tagged in Fall 2019 to an individual that measured 171 millimeters tagged in Fall 2018.

## Demographic Rate Estimation

A total of 7,140 Brook Trout were estimated to be residing in Tyee Springs in fall 2019 (Table 2). Abundance by age class was estimated at 4020 age zero Brook Trout, 2744 age one Brook Trout and 376 for ages two and older (Table 3). Stage-specific survival rate and fertility rate estimates are in Table 3. Ninety-five percent credible intervals were calculated for both abundance and survival rate estimates; these were used to inform low and high values for the one-way sensitivity analysis. A reasonable range of values were chosen to explain fertility rate uncertainty for the one-way sensitivity analysis.

Table 2. Abundance estimates with $\mathbf{9 5 \%}$ credible intervals for three sampling periods (fall 2018, spring 2019, and fall 2019) by sampling unit.

|  | Units | $2.5 \%$ | $50 \%$ | $97.5 \%$ |
| :--- | :---: | :---: | :---: | :---: |
| Fall 2018 | 1 | 236 | 288 | 364 |
|  | 2 | 904 | 2,459 | 10,844 |
|  | 3 | 2,374 | 3,850 | 6,561 |
|  | 1 | 123 | 155 | 208 |
| Fall 2019 | 2 | 547 | 1,768 | 12,142 |
|  | 3 | 646 | 1,037 | 1,858 |
| Fall 2018 | 1 | 311 | 404 | 549 |
| Spring 2019 | 2 | 614 | 954 | 1,631 |
| Fall 2019 | 3 | 3,805 | 5,749 | 9,257 |

Table 3. Parameters used to populate population matrix model and associated 95\% credible intervals, where $N_{0}=$ starting abundance, $S=$ survival rates and $F=$ Fertility rates.

|  | $\mathrm{N}_{0}$ | $95 \% \mathrm{CIs}$ | S | $95 \% \mathrm{CIs}$ | F | $*$ |
| :--- | :---: | :---: | :---: | :---: | :---: | :---: |
| Age-0 | 4020 | $2569-6685$ | 0.44 | $.28-.66$ | 0 | - |
| Age-1 | 2744 | $2086-3797$ | 0.14 | $.09-.23$ | 2.5 | $1.1-5$ |
| Age-2+ | 376 | $288-513$ | 0.08 | $.05-.13$ | 7.2 | $3.6-14.2$ |

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## Model simulations

When annual fish suppression is held at $50 \%$ (as recommended by Schill et al. 2017) and $\mathrm{M}_{\mathrm{yy}}$ stocking is set at a constant 3,000 per year, the model estimates that eradication will occur on average nine years after initial stocking of $\mathrm{M}_{\mathrm{yy}}$ Brook Trout with an $80 \%$ probability that it will be between seven and fourteen years due to demographic stochasticity. When annual suppression rates are lowered to $25 \%$, the estimated number of years until eradication increases to thirteen with an $80 \%$ of stochastic simulations falling between nine and nineteen years (Figure 8). Given that we believe our suppression rates will likely decline over time as available fish abundance decreases, we also tested suppression rate as a function of abundance. When fish suppression is set at $50 \%$, but slowly decreases to zero when fish abundance drops to 300 , and the annual stocking rate is set at 3,000 , our estimated number of years until eradication is thirteen versus nine years when suppression rate was held at a constant $50 \%$ (Figure 8).

## Sensitivity Analyses

One-way sensitivity analyses identified annual suppression rate as the most influential model parameter (Figure 9). When fish suppression is held at a very low rate of $5 \%$, we would expect to see eradication occur in 18 years on average after first stocking Myy Brook Trout when all other parameters are held at their initial values. Conversely, when annual suppression is held at $95 \%$, we would expect eradication to occur in four years. The next three most sensitive parameters were age- 0 survival, age- 1 fertility rate, and age- $2+$ fertility rate, respectively.

Annual rate of stocking is the fifth most sensitive model parameter, only varying the outcome from eight to ten years. However, this is partly due to the suppression rate being fixed at $50 \%$ during the one-way sensitivity analysis; the stocking parameter became increasingly more sensitive as suppression rates decreased. This relationship can be seen in the two-way sensitivity output (Figure 10). As suppression rates are increased, the number of years until eradication decreases at a high rate given any reasonable value for the stocking rate parameter. In contrast, as stocking rates are increased, time to eradication decreases significantly only when suppression
rates are low. Given that we are uncertain how effective our suppression efforts of the natural population will be at this time, the two-way sensitivity output can help guide where to focus our efforts as well as inform eradication timing.

## Discussion

Nonnative Brook Trout pose a health risk to juvenile spring Chinook reared at Carson NFH and a potential ecological risk to native fish species in the Wind River and ESA-listed bull trout in the Walla Walla River. Many of the control methods used to eradicate unwanted fish populations are cost and/or labor intensive (e.g., harvest or physical removal), harmful to the aquatic ecosystem (e.g., piscicides) and vary widely in their effectiveness. The $\mathrm{M}_{\mathrm{yy}}$ technique (i.e., skewing the sex ratio of the resident population) may be an optimal fish control strategy for Tyee Springs because it's a small isolated stream with no recruitment from outside populations (i.e., Wind River), Brook Trout and sculpin are the only fish species that currently occupy Tyee springs (i.e., limited competition for resources), $\mathrm{M}_{\mathrm{yy}}$ Brook Trout pose no additional risk to spring Chinook at Carson NFH, $\mathrm{M}_{\mathrm{yy}}$ fish are readily available for stocking, and the technique has shown promising preliminary results in the four western states currently stocking $\mathrm{M}_{\mathrm{yy}}$ Brook Trout (Kennedy et al 2018; Roth et al. 2019).

The purpose of our study was to determine the baseline population structure and abundance of resident Brook Trout inhabiting Tyee Springs in order to perform a population modeling exercise to evaluate the feasibility of stocking $\mathrm{M}_{\mathrm{yy}}$ fish to eradicate nonnative Brook Trout in Tyee Springs. A secondary goal of the simulation exercise was to identify optimal fish suppression and/or $\mathrm{M}_{\mathrm{yy}}$ stocking rates that would lead to successful eradication in a reasonable time frame. From a management perspective, time to eradication is one of the most important factors in the decision to implement the $\mathrm{M}_{\mathrm{yy}}$ approach. Results from our modeling work indicate that not only is eradication of nonnative Brook Trout possible in Tyee Springs, but time to eradication generally decreases as annual fish suppression rates are increased. This is consistent with previous work demonstrating that annual suppression of the resident population is necessary to shorten time to eradication (Day et al. 2020; Schill et al. 2017). For instance, in a simulation scenario where we stock 3,000 Myy Brook Trout annually ( $\approx 50 \%$ of resident Brook Trout population) and maintain a suppression rate of $50 \%$ annually, 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. We consider around 10 years to be a reasonable time frame to achieve fish eradication, but this is dependent upon our ability to mechanically remove at least half of the resident Brook Trout population in Tyee Springs annually. Maintaining an annual suppression rate of $50 \%$ will likely be difficult given suppression rates will decline over time as resident fish abundance decreases. To model this scenario, we tested suppression rate as a function of abundance, incrementally reducing the suppression rate as resident Brook Trout
abundance declines. Under these conditions, the estimated number of years to eradication increased to 13 years versus 9 years when suppression was held at a constant $50 \%$. Achieving a $50 \%$ suppression rate will require extensive fishing effort over multiple days or weeks. A possible alternative to expending more effort to suppress the resident population would be to increase $\mathrm{M}_{\mathrm{yy}}$ stocking rates. However, Our sensitivity analyses indicates that there is no difference in time to eradication between stocking 3,000 or $7,000 \mathrm{M}_{\mathrm{yy}}$ Brook Trout at suppression rates $\geq 25 \%$ (see Figure 10). Rather than expend additional time and resources to rear more $\mathrm{M}_{\mathrm{yy}}$ Brook Trout, we propose stocking $\mathrm{M}_{\mathrm{yy}}$ fish at a rate equivalent to $50 \%$ of the total resident Brook Trout population and employing multiple fish removal methods over several days to maximize annual suppression rates to the greatest extent possible.

Field implementation of the Myy approach is relatively new and assumptions were made in the population simulation model that likely influenced our brook trout eradication projections. First, we assumed Brook Trout suppression rates were constant across all age classes. After implementing suppression efforts in Tyee Springs, we may observe bias related to our capture methods and fish size. In future revisions of the model, suppression rates will be inferred from rates observed during fish removal efforts. Next, we assumed $\mathrm{M}_{\mathrm{yy}}$ survival post-stocking/prespawning to be 18 percent and annual survival to be 5 percent. Preliminary data from field studies have estimated annual survival of stocked $\mathrm{M}_{\mathrm{yy}}$ brook trout between $3 \%$ and $18 \%$. We also assumed that stocked $\mathrm{M}_{\mathrm{yy}}$ fish were equally fit to spawn (i.e., reproduced as effectively) as naturally produced adult males. We elected to parametrize the model with conservatively low survival rates and assume zero difference in spawning fitness (between resident and $\mathrm{M}_{\mathrm{yy}}$ populations) until these metrics can be assessed in Tyee Springs. Demographic parameters may be higher or lower than our current assumptions, which is why long term monitoring of the population will be an essential component of our study. We recommend implementing an adaptive management approach, using annual fish removal events as an opportunity to monitor the population and collect additional information to update and improve the population model. Some additional monitoring may include:

- Assessing the efficiency of our population suppression techniques, associated variance and how our efficiency is affected by the decline in abundance.
- Using genetic parentage analysis to determine what proportion of Brook Trout fry are progeny of an $\mathrm{M}_{\mathrm{yy}}$ parent (i.e., spawning success).
- Estimating the survival of stocked $\mathrm{M}_{\mathrm{yy}}$ fish.
- Assessing fish removal capture rates by age class.
- Monitoring female abundance and/or sex ratio through time in an effort to determine eradication timing.

We plan to draft a study plan that clearly defines our future $\mathrm{M}_{\mathrm{yy}}$ monitoring strategy with the goal of maximizing eradication and informing future population modeling efforts.

The FDX PIT tag antenna array detected a total 24 tagged Brook Trout leaving Tyee Springs during the roughly six months it was operational. This low level of emigration is somewhat concerning, given the potential for $\mathrm{M}_{\mathrm{yy}}$ Brook Trout to enter the Wind River where they could pose a threat to native fish populations. Within the scope of this study, it is also possible that $\mathrm{M}_{\mathrm{yy}}$ Brook Trout might emigrate at even higher rates than the resident population, thereby extending our time to eradication. To reduce the potential risk associated with $\mathrm{M}_{\mathrm{yy}}$ Brook Trout emigration to the Wind River, the Columbia River FWCO and Carson NFH are working with Washington Department of Fish and Wildlife fish screen experts to fabricate and install a screening system in the bypass channel that will act as a complete passage barrier to Brook Trout. The screen will be installed before a large scale stocking effort is implemented in Tyee Springs. The majority of current $\mathrm{M}_{\mathrm{yy}}$ field studies are stocking $\mathrm{M}_{\mathrm{yy}}$ Brook Trout in 'open' streams where fish may move freely to other locations. Even though an unknown percentage of $\mathrm{M}_{\mathrm{yy}}$ fish may leave the target stream, these projects are still observing beneficial results (i.e., detection of $\mathrm{M}_{\mathrm{yy}}$ progeny). After the screening system is installed and Tyee Springs is a completely closed system, we expect to see even higher rates of $\mathrm{M}_{\mathrm{yy}}$ introgression than those observed in other $\mathrm{M}_{\mathrm{yy}}$ field evaluations; potentially reducing our time to eradication.

If the eradication of Brook Trout is successful in Tyee Springs, the Service will ultimately save money by eliminating the cost of maintaining the physical fish screens and having to manually sort hatchery fish prior to release. Successful eradication will also remove the threat of inadvertently releasing Brook Trout into habitat occupied by native ESA-listed bull trout and will eliminate a vector for BKD transmission to Chinook salmon in Carson NFH. This proof-ofconcept evaluation can help the Service and its partners determine if and how the $\mathrm{M}_{\mathrm{yy}}$ 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 aquatic invasive species, like common carp, which have proven very difficult to control in places like Malheur National Wildlife Refuge. Finally, this will be one of the first studies to assess how well the simulation model predicts the time to eradication of a resident Brook Trout population in a natural stream using the Trojan Y Chromosome technique.

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Figure 1. Map of Carson National Fish Hatchery and Tyee Springs showing three survey units, 2019.


Figure 2. Sampling Unit 1: hatchery intake channel.


Figure 3. Sampling Unit 2: free flowing stream.


Figure 4. Sampling Unit 3: headwaters of Tyee Springs.


Figure 5. FDX PIT tag antenna array located in secondary bypass channel, immediately downstream from sampling Unit 1.


Figure 6. Diagram of stage based population model structure, where Sage indicates age specific survival rates and $F_{\text {age }}$ indicates age specific fertility rates.


Figure 7. Bypass Channel Detections and average precipitation (in) per week from October 21, 2019 to March 12, 2020.


Figure 8. Two bar plots representing stochastic simulation outputs ( $\mathbf{N}=1000$ ) when annual stocking rates are held at $\mathbf{3 0 0 0}$, and annual suppression rates are held at $\mathbf{5 0 \%}$ (top) and 25\% (bottom).


Figure 9. One-way sensitivity analysis tornado plot. One model parameter was varied from reasonable low to high values while all other parameters were held at their mean value. The estimated number of years until eradication occurs was recorded for every parameter perturbation.


Figure 10. Two-way sensitivity analysis results. The number inside each cell represents the estimated number of years until eradication occurs given changes to Myy stocking and suppression parameters.

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[^0]:    * As fecundity rate credible intervals were not calculated, plausible values were chosen for the one-way sensitivity analysis based on local knowledge and literature review

