

Estuary Entry and Residency Patterns Amongst Summer Run Puntledge Chinook Salmon

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Salmon

By

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ABSTRACT

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Sagittal otoliths from 32 natural origin and 91 hatchery origin summer run Puntledge River Chinook salmon (*Oncorhynchus tshawytscha*) were collected in the 2021 return year from the hatchery broodstock collections. Of these otoliths, 32 natural origin and 68 hatchery origin Chinook otoliths were selected to be analysed by Laser Ablation Inductively Coupled Plasma Mass Spectrometry (LA-ICP-MS) to identify the outmigration behaviours of these Chinook salmon. While hatchery and natural origin Chinook salmon entered the estuary at the same time, natural origin Chinook were more likely to use the estuarine habitat for great lengths of time, especially amongst the smallest outmigrants. 34% of the natural origin Chinook and 1% of the hatchery origin Chinook showed evidence of extensive estuary usage. 94% of the natural origin Chinook appeared to use the lower river habitat for a significant amount of time after making it beyond the Puntledge River Diversion Dam. Natural origin Chinook displayed a greater range in phenotypic diversity in terms of habitat usage, migration patterns, and age at return. Considerations of genotypic and phenotypic diversity amongst summer run Puntledge River Chinook should be engaged in further discussions for the continued survival and enhancement of this stock.

RÉSUMÉ

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Des otolithes sagittaux de 32 saumons Chinook (*Oncorhynchus tshawytscha*) d'origine naturelle et 91 d'écloserie remontant la rivière Puntledge durant l'été 2021 ont été collectés parmi le stock de reproducteurs de l'écloserie. Parmi ces otolithes de saumons Chinook, 32 d'origine naturelle et 68 d'écloserie ont été sélectionnés pour être analysés par spectrométrie de masse à plasma à couplage inductif par ablation laser (LA-ICP-MS) afin d'identifier les comportements de migration de ces saumons. Bien que les saumons d'origine naturelle et ceux d'écloserie soient entrés dans l'estuaire en même temps, les saumons d'origine naturelle étaient beaucoup plus susceptibles d'utiliser l'habitat estuarien pendant de longues périodes, en particulier les plus petits migrants. 34% des saumons d'origine naturelle et 1 % des saumons d'écloserie ont montré des preuves d'utilisation de l'estuaire. 94% semblaient utiliser l'habitat de la rivière inférieure pendant une période significative après avoir passé le barrage de diversion de la rivière Puntledge. Les saumons Chinook d'origine naturelle ont montré une plus grande diversité phénotypique en termes d'utilisation de l'habitat, de motifs de migrations et d'âge au retour. Les diversités génotypique et phénotypique des saumons Chinook qui remontent la rivière Puntledge en été devraient être prises en compte dans les discussions ultérieures pour la survie continue et l'amélioration de cette population.

1. INTRODUCTION

The Puntledge River Watershed is one of the few East Coast Vancouver Island (ECVI) river systems that contains runs of both summer and fall run Chinook salmon (Guimond & Withler, 2010). Historically, the Puntledge River system saw significant production and was one of 15 streams that were known to have adult spawning returns over 5,000 individuals (DFO, 1958; Trites et al., 1996). Roughly half of these Chinook salmon were summer run, and they typically spawned between the site of the Puntledge River Diversion Dam and Comox Lake (MacKinnon et al., 1979). These Chinook entered the river system from June-August and were able to ascend the Stotan and Nib Falls which were less accessible to fall run Chinook later in the season (MacKinnon et al., 1979; Trites et al., 1996). Summer run Puntledge (SPUN) Chinook may have evolved from the early return migrants of fall run Chinook through a parapatric process, with Stotan and Nib Falls acting as a reproductive barrier. Some mixed fish likely still existed due to some overlap in return migrations.

As adults, SPUN Chinook face numerous threats from hydroelectric activities, seal predation within the river and nearshore environment, and angler capture (DFO, 1958; MacKinnon et al., 1979; Trites et al., 1996). These factors have culminated in an escapement level that is consistently $1/10^{\text{th}}$ the historical escapement despite 60 years of considerable efforts (Guimond & Taylor, 2017; MacKinnon et al., 1979; Trites et al., 1996). This population was declared endangered in 2020 by COSEWIC as part of Designatable Unit 20 (ECVI, Ocean, Summer run Chinook). While removing barriers to egg to smolt survival through hatchery production has been one point of research for this population, there is interest in understanding and improving conditions for the out-migrating natural origin juveniles through habitat restoration efforts.

Chinook salmon demonstrate a wide variety of life history strategies, especially during their juvenile life stage. They can spend weeks to over a year in the freshwater before their downstream migration, and this can be influenced by a wide variety of endogenous and exogenous factors (Beckman et al., 1998; Bourret et al., 2016; Healey, 1991; Quinn et al., 2005). Within SPUN Chinook, natural origin juveniles are subyearling type Chinook, with most out-migrating from February until July (Lister, 1968; Trites et al., 1996). There appears to be a bimodal pattern in outmigration with a numerous <50mm cohort leaving for the estuary between the end of February to the beginning of May, and a small >60mm cohort leaving for the estuary around June (Guimond & Taylor, 2015; Lister, 1968). Hatchery origin Chinook, by comparison, are all released in late May at >65mm and >3.0g. This lack of life history diversity in hatchery Chinook in general is paired with increased stressors on these smolts, such as increased likelihood of vaterite formation, and poorer overall survival and fecundity (Beamish et al., 2012; Bourett et al., 2016; Gaffney et al., 2025). Emergent fry below 50mm can make up over half of the total downstream migrators amongst natural origin SPUN Chinook, but these suffer higher rates of entrainment mortality due to screens meant to divert fry from the dam (Guimond & Taylor, 2017). Natural origin Chinook make up only a small proportion of SPUN Chinook returns, likely due to this high early mortality, but are critical to providing additional genetic diversity to the overall population.

One aspect of these Chinook that is poorly understood is the use of the estuarine environment and how it may differ between these three types of smolts (emergent fry, parr, and hatchery out-migrants). Subyearling type Chinook salmon rely on the estuarine and nearshore marine environments to accelerate their growth (Chalifour et al., 2021). Chinook salmon in the Nanaimo River, one of the other mixed summer and fall run groups on the east coast of Vancouver Island, were found to grow over 1mm a day in the estuarine environment, indicating it is an important habitat for smolt rearing (Healey, 1971). Differences in estuary use amongst returning adults may explain some of the trends in smolt to adult survival between natural origin and hatchery origin SPUN Chinook.

In this study, we have investigated estuary use and residence differences between natural and hatchery origin SPUN Chinook for the 2021 brood year. We used Laser Ablation Inductively Coupled Plasma Mass Spectrometry (LA-ICP-MS) to identify the intrinsic elemental markers within otoliths that are related to habitat change. Strontium concentrations in otoliths correlate with water concentrations, as calcium within the aragonite lattice substitutes with strontium more frequently as strontium concentrations increase (Thomas & Swearer, 2019; Tripp et al., 2022). Since strontium is an intrinsic part of the salt concentrations within seawater, overall strontium concentrations increase with salinity, and thus this can show transitions from freshwater to saltwater (Kalish, 1989). We conducted line ablations from the core to the edge of the otolith for 100 SPUN Chinook escapements. Our goals were to: 1) identify the proportion of early and late migrants amongst natural origin Chinook; and 2) identify estuary residence patterns between early and late natural origin migrants, and hatchery migrants.

2. METHODS

2.1 SAMPLE COLLECTIONS

There were 199 Chinook salmon collected from the returning spawners in 2021 all of which were sampled for genetics to determine their origin. 109 of these Chinook salmon had scale ages and 186 had genetic based ages. Genetic based ages were treated as the most accurate, with scales supplementing ages where brood year was undeterminable from the genetic analyses. 123 SPUN Chinook salmon had their otoliths extracted for this study. Of these, 97 were spawned as part of hatchery enhancement efforts.

2.2 OTOLITH MEASUREMENTS

Otoliths were briefly submerged (maximum 3 min) in a plastic Petri dish filled with Super-Q deionized water (MilliporeSigma, Burlington, MA, USA; www.emdmillipore.com). The distal sides of the otoliths were photographed on a black background using an Olympus SZX16 stereoscope, DP26 camera and CellSens Standard software (Olympus, Shinjuku, Tokyo; www.olympus-global.com) at 16× magnification (Figure 1a). Adobe Photoshop 2020 (Adobe

Incorporated, San Jose, CA, USA) was used to modify the otolith photos to make the otolith appear pure white (hexidecimal value #FFFFFF) and the background pure black (hexidecimal value #000000; Fig 1b). This was done to conduct brief morphometric assessments on the otoliths. Adobe Photoshop 2020 (Adobe Incorporated, www.adobe.com) was used to differentially colour pixels that represented aragonite and vaterite as determined by an experienced technician. Aragonite was coloured in pure red (hexidecimal value #FF0000), and vaterite was coloured in pure green (hexidecimal value #00FF00; Fig 1c). Otoliths with more than 5% vaterite incorporation were considered to be vateritic.

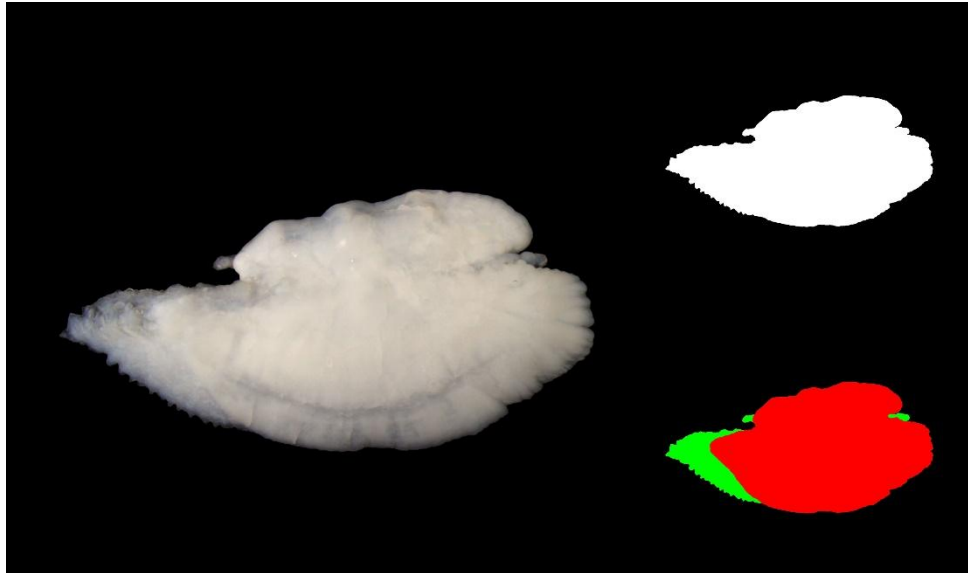


Figure 1. Comparison of otolith views for the left otolith of SPUN21-20. 1A: The natural photograph of the otolith without any modification. 1B: The outline of the otolith where the entire otolith is highlighted white to aid shapeR in automatic outline detection. 1C: The green areas represent vaterite (or calcite) while the red areas represent aragonite which is the natural calcium carbonate polymorph in otoliths.

2.3 OTOLITH RADIUS TO FORK LENGTH RELATIONSHIP

To relate otolith radius to fork length, two different measurements were taken from 43 otoliths collected from juvenile summer run Puntledge Chinook salmon measuring from 35 to 81mm (Figure 2). The first was the radius from the core of the otolith to the postrostrum (located in the posterior and ventral quadrant of the otolith). The second measurement was from the otolith core to a 20-degree angle posterior of the dorsal midline of the otolith. These Chinook juveniles were collected from regular health checks from the hatchery population between February to June of 2024. The fork lengths of these fish were collected in field soon after capture. Measurements were made in the CellSens Standard software (Olympus, Shinjuku, Tokyo; www.olympus-global.com).

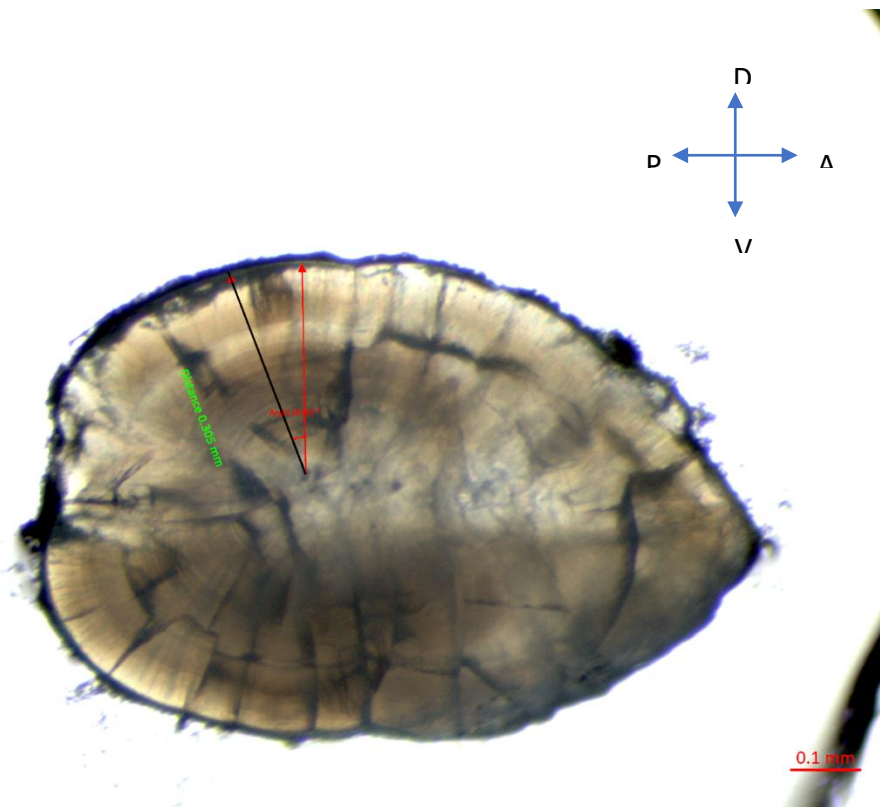


Figure 2. Otolith from an 42mm summer run Puntledge Chinook salmon (*Oncorhynchus tshawytscha*) showing the line drawn from the core of the otolith to the postrostrum edge of the otolith as well as the line drawn from the core to a point 20 degrees posterior of the dorsal midline of the otolith. The scale bar represents 100 μm . The most posterior primordia was always selected in every individual. Compass identifies the orientation in the otolith, with the dorsal (D), ventral (V), posterior (P), and anterior (A) quadrants labelled. The scale bar represents 100 μm .

2.4 OTOLITH MICROCHEMISTRY ANALYSES

After otoliths had been photographed, otoliths were sub-sampled for elemental analysis. Unbroken aragonitic otoliths were selected from fish. The left otolith was selected preferentially to avoid any error due to population level asymmetries. Otoliths from spawned individuals were preferentially selected, with 100 individuals in total selected for trace element analysis. These otoliths were placed sulcus up within 15mmx15mm Fisherbrand Disposable Base Molds (Thermo Fisher Scientific, Waltham, MA, USA; www.thermofisher.com). To produce a flat surface for later sanding, the rostrum was generally removed to allow the otolith to lay flat. EpoxiCure 2 (Buehler, Lake Bluff, IL, USA; www.buehler.com) was poured onto the otoliths to solidify them within an epoxy block. After 24 hours, epoxy blocks were sanded down using progressively finer sandpapers (320, 600, 1200 grit sizes) to expose the core of the otolith.

Laser line ablations were conducted from just before the core of the otolith to the post-rostrum edge. The LSX-213 G2+ Nd:YAG laser ablation system (Teledyne Technologies,

Thousand Oaks, CA, USA; www.teledyne.com) was used in concert with an Agilent 8800 ICP-MS system (Agilent, Santa Clara, CA, USA; www.agilent.com). The laser spot size was 30µm, the laser speed was 10µm/sec, and the element dwell time was 10 ms. A ~25 second gas blank baseline before and after the ablation pattern was conducted for each otolith. Six elements in total were measured: ^{25}Mg , ^{43}Ca , ^{55}Mn , ^{66}Zn , ^{88}Sr , and ^{138}Ba . NIST glass standards 610, 612, and 614 (National Institute of Standards and Technology, Gaithersburg, MD, USA; www.nist.gov) were used as external standards. Calcium was assumed to be at 40.04% by weight in the otoliths, and thus this was used as the internal standard. Iolite 4 (iolite.xyz) was used for data reduction and processing. The otolith core, estuary entry, and full oceanic transition were determined through expert determination aided by the absolute and relative to calcium concentrations of the various elements within the otolith.

2.5 ESTUARY RESIDENCY ANALYSIS

Along with juvenile and adult Chinook salmon otoliths, ten otolith pairs were collected from Chinook salmon smolts collected from the Puntledge River estuary in early June of 2021. Genetics were not run on these salmon, so they could be summer or fall run individuals. One otolith (the right) was prepared for laser line ablations while the other (the left) was prepared for daily ring measurements. These otoliths were prepared in a similar manner as described in Chalifour et al., 2021. The daily growth rate in the estuary was measured by averaging the individual distances between daily rings over the total number of rings present during their estuary entry period. Daily rings were counted and measured in situ using an Olympus BX53 compound microscope along the 20-degrees axis described above. We assumed estuary entry occurred where the strontium incorporation signal began, though this was usually near a check mark visible in the otolith microstructure. We assumed entry into the marine environment when the Ba/Sr matched samples known to have resided in the Strait of Georgia. The difference in these two locations on the otolith were then divided by the average circuli spacing related to estuarine growth. Those individuals that resided in the estuary for more than the typical transit and smoltification time (> two weeks) were considered to be rearing in the estuarine environment (Greene et al., 2025).



Figure 3. Otolith of a natural origin Chinook salmon (*Oncorhynchus tshawytscha*) captured in the Courtney River estuary prepared to measure the distance between daily rings. This individual had been in the estuary for 62 days, entering on April 9th, 2021 and being captured on June 10th, 2021. The strontium inflection point occurred at the distance indicated by the red arrow when the fish was predicted to be 36mm.

2.6 STATISTICAL ANALYSES

Permutation Welch's two sample t-tests were used to determine differences in post-orbit to hypural plate (POH) lengths, estimated estuary entry fork length, and estimated ocean entry fork length between hatchery and natural origin SPUN Chinook. 9999 Monte Carlo permutations were conducted for all permutation t-tests. Chi-square tests were used to identify differences in vaterite prevalence between SPUN origin, age, and sex. Cochran-Mantel-Haenszel chi-squared tests for count data were conducted to test the hypothesis of whether AA, AV, and VV otolith pairs were equally represented in both hatchery and natural origin Chinook across sexes and ages (Cochran, 1954; Mantel, 1963).

3. RESULTS

3.1 SIZE AND AGE COMPOSITION OF HATCHERY AND NATURAL ORIGIN CHINOOK

In total, there were 165 hatchery origin SPUN Chinook and 33 natural origin Chinook salmon collected for SPUN broodstock. One salmon was from the Nanaimo River, but its origin was unresolved and thus removed from further analysis. Two other strays were included, one hatchery origin salmon from the fall run in the Puntledge River, and one hatchery origin salmon from the summer run Nanaimo River population. Their inclusion did not impact any of the results reported below. Natural origin Chinook salmon averaged at 604mm from post-orbit to hypural plate (POH), whereas hatchery origin salmon averaged at 535mm (Fig. 4). This difference was significant ($p = 0.004$) likely due to the greater frequency of occurrence of four-year-olds in the natural origin population ($\chi^2 = 23.179$, $df = 3$, $p < 0.001$; Fig. 5). This pattern holds true if scale age is used as well, though the five-year-old in the natural origin component of the population is now a four-year-old ($\chi^2 = 11.523$, $df = 2$, $p = 0.003$). Size at age was not significantly different between natural origin and hatchery origin Chinook salmon using the genetic based age ($F = 0.240$, $df = 1$, $p = 0.664$; Fig. 6), though it was significant when using the scale-based age ($F = 3.979$, $df = 1$, $p = 0.047$). The average age of hatchery origin Chinook salmon (3.2) was lower than natural origin (3.6) component of the population ($p = 0.012$). The median age for hatchery origin fish was three, while the median age for natural origin fish was four.

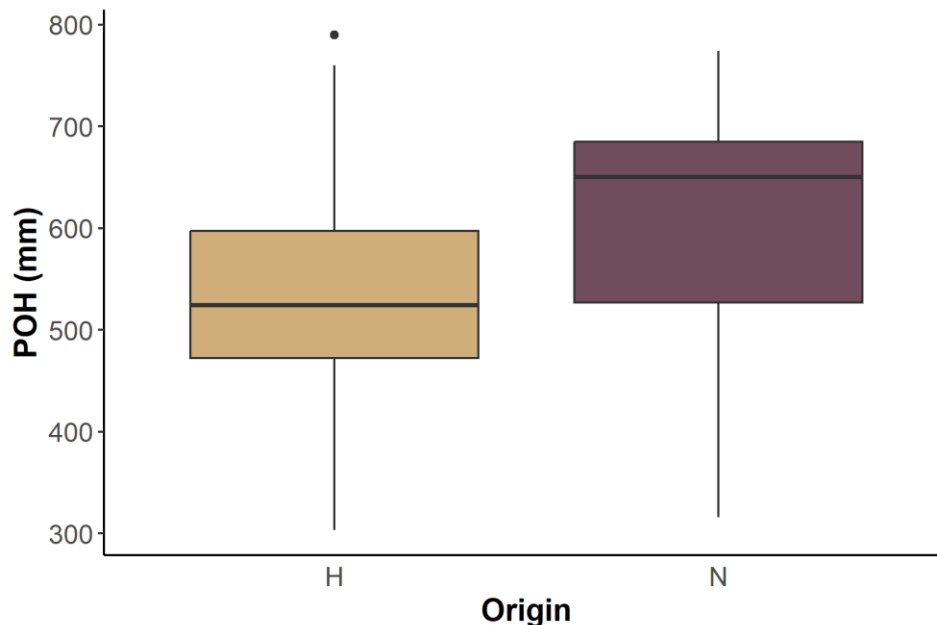


Figure 4. Boxplot showing the difference in post-orbit to hypural plate (POH) lengths between hatchery origin (gold) and natural origin (purple) summer run Puntledge Chinook salmon (*Oncorhynchus tshawytscha*). $n = 165$ for hatchery origin fish and $n = 33$ for natural origin fish. Results include two hatchery strays, one from fall run Chinook salmon of Puntledge River and one from the summer run Chinook salmon of Nanaimo River.

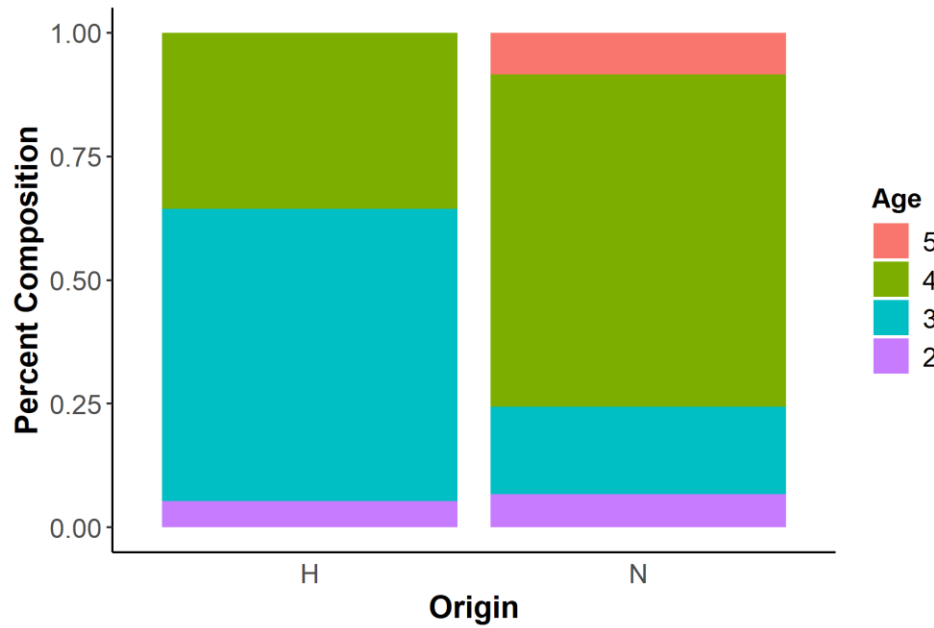


Figure 5. Stacked bar plot showing the differences in age compositions between hatchery origin and natural origin summer Puntledge Chinook Salmon. The median age was 4 for each group. The mean age was 3.19 for hatchery origin and 3.55 for natural origin Chinook salmon. Results include two hatchery strays, one from a fall run Chinook salmon of Puntledge River and one from a summer run Chinook salmon of Nanaimo River. $n = 165$ for hatchery origin fish and $n = 33$ for natural origin fish. Minijacks were not included.

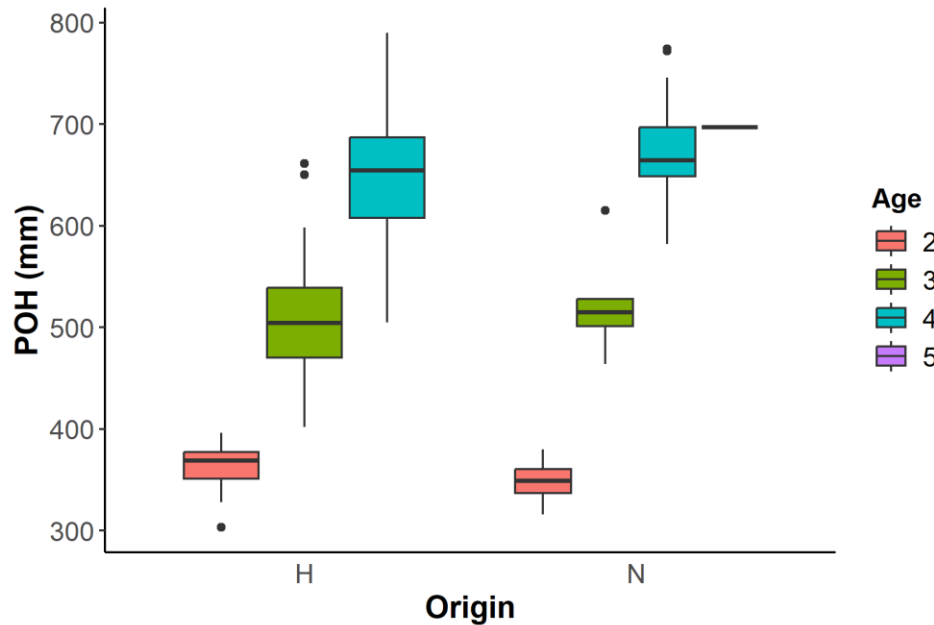


Figure 6. Boxplot showing the difference in post-orbit to hypural plate (POH) lengths between hatchery origin (H) and natural origin (N) summer run Puntledge Chinook salmon (*Oncorhynchus tshawytscha*). n = 165 for hatchery origin fish and n = 33 for natural origin fish. Results include two hatchery strays, one from fall run Chinook salmon of Puntledge River and one from the summer run Chinook salmon of Nanaimo River.

3.2 VATERITE MEASUREMENTS

26% of the otoliths surpassed the 5% vaterite threshold and were labeled vateritic as a result. This amounted to 47 of the 123 summer Puntledge Chinook salmon containing at least one vateritic otolith (Table 1). Vaterite was far more likely to occur in hatchery origin fish ($\chi^2 = 18.964$, $df = 2$, $p < 0.001$). Vaterite frequency was not different between males and females ($\chi^2 = 1.708$, $df = 2$, $p = 0.426$), nor between different age classes ($\chi^2 = 4.121$, $df = 6$, $p = 0.660$). When accounting for age, vaterite frequency was significantly higher amongst hatchery origin Chinook ($M^2 = 16.694$, $df = 2$, $p < 0.001$). The greatest difference was between 4-year-old hatchery and natural origin chinook ($p_{adj} = 0.005$). When accounting for sex, vaterite frequency was significantly higher amongst hatchery origin Chinook ($M^2 = 17.977$, $df = 2$, $p < 0.001$). The greatest difference was between female hatchery and natural origin chinook ($p_{adj} = 0.007$). In AV pairs, vaterite was far more likely to occur in the left rather than the right otolith ($\chi^2 = 8.067$, $df = 1$, $p = 0.005$).

Table 1. Number of natural and hatchery origin SPUN Chinook that have aragonite-aragonite (AA), aragonite-vaterite (AV), and vaterite-vaterite (VV) pairs.

	AA	AV	VV	Total
Natural Origin	30	2	0	32
Hatchery Origin	46	28	17	91
Total	76	30	17	123

3.3 OTOLITH RADIUS TO FORK LENGTH RELATIONSHIP

Of the 43 juvenile otoliths, 40 were able to be measured as three contained significant amounts of vaterite. The linear regression between fork length and the postrostrum otolith radius did meet the assumptions of a linear relationship, and this relationship ($y = 0.104 * x + 4.194$) had a high adjusted R^2 value at 0.841 (Fig. 7). We also modelled the fork length to the 20 degrees posterior to the dorsal midline and found this relationship ($y = 0.188 * x - 18.060$) to have an even higher adjusted R^2 value at 0.912 (Fig. 8). While breakpoint regressions have been useful to measure the fork lengths of natural origin populations, the regression from the hatchery origin juveniles lacked a clear shift in growth between the fry and parr stage shift which occurs around 45mm. Therefore, a simple linear regression was used, though this may slightly overestimate the fork length of natural origin Chinook salmon that entered the estuary as fry.

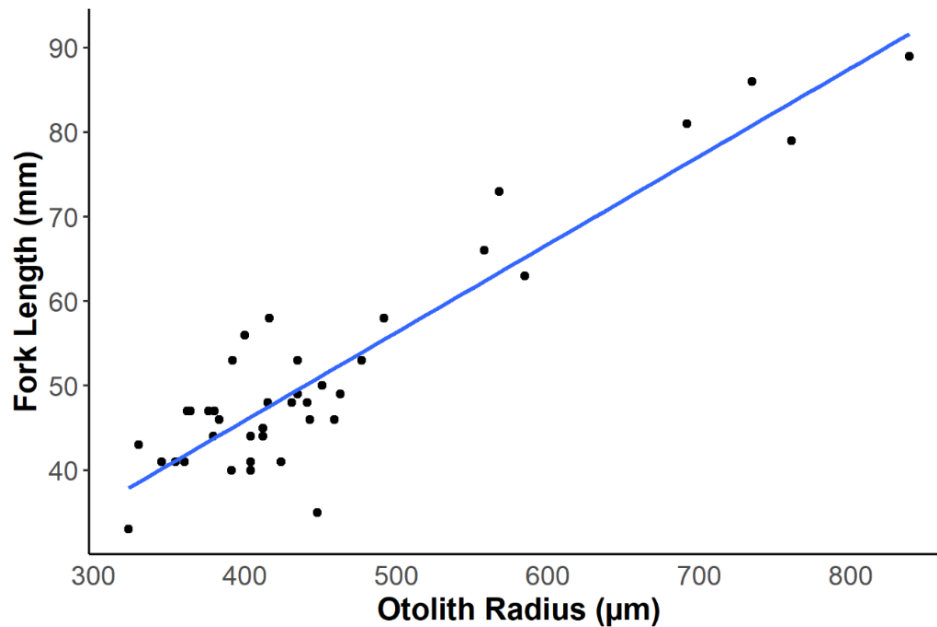


Figure 7. Linear relationship between the otolith radius from the primordia to the postrostrum and the fork length of juvenile summer Puntledge Chinook salmon (*Oncorhynchus tshawytscha*) collected from the Puntledge River Hatchery in 2024. The equation is $y = 0.104 * x + 4.194$ with an R^2_{adj} of 0.841. $n = 40$.

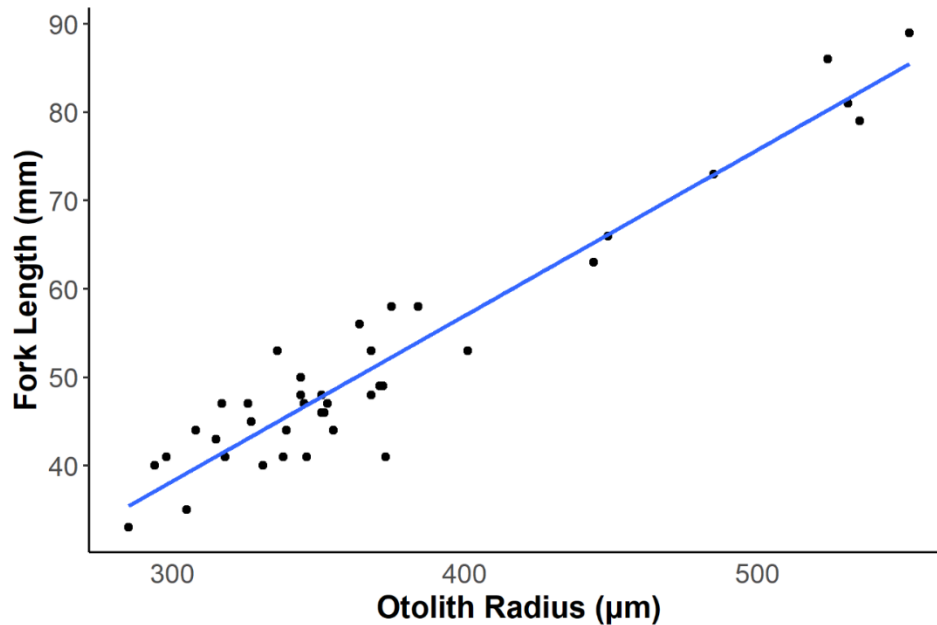


Figure 8. Linear relationship between the otolith radius from the primordia to a point on the otolith edge 20 degrees to the posterior of the dorsal midline and the fork length of juvenile summer Puntledge Chinook salmon (*Oncorhynchus tshawytscha*) collected from the Puntledge River Hatchery in 2024. The equation is $y = 0.188x - 18.060$ with an R^2_{adj} of 0.912. $n = 40$.

3.4 ESTUARY ENTRY AND RESIDENCY

Only 100 otoliths were run for LA-ICP-MS analyses. On average, natural origin Chinook entered the estuary at a larger size (76.2mm) than hatchery origin Chinook (72.3mm), but this difference was not significant ($p = 0.451$; Fig 9). Natural origin Chinook entered the estuary across a much broader range of sizes (34.0 – 162.0mm) than hatchery origin Chinook (37.3 – 85.4 mm). When breaking these into life history types (30-50mm, 51-70mm, and >70mm estuary entry types), the vast majority of Puntledge summer Chinook were represented by the largest (>70mm) life history type (Table 2). The three largest natural origin Chinook salmon leaving the river system corresponded to individuals that were estimated to be older according to their genetic based age (based on when candidate parents returned to the river to spawn) than their scale-based age. Estuary entry life-history types differed significantly between natural and hatchery origin Chinook ($\chi^2 = 10.162$, $df = 2$, $p < 0.007$). Hatchery origin Chinook were composed primarily of the >70mm life-history type, while most of the Chinook in the <50mm life-history type were of natural origin. Several natural origin Puntledge summer Chinook entered the estuary at over 100mm, which could indicate the presence of yearlings in the Puntledge River.

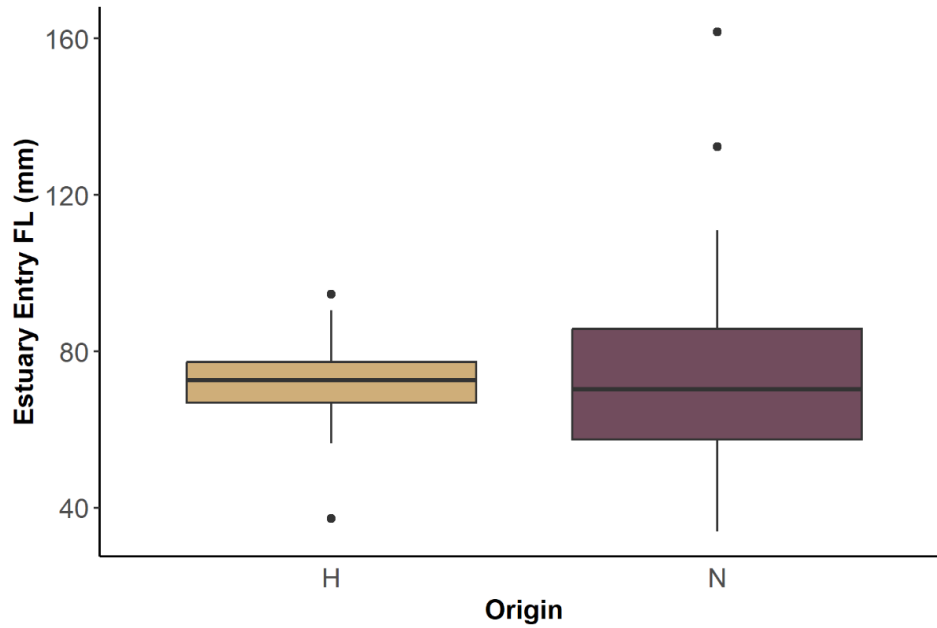


Figure 9. Predicted estuary entry fork lengths for hatchery (gold) and natural origin (purple) summer Puntledge Chinook. $n = 68$ for hatchery origin fish and $n = 32$ for natural origin fish.

Table 2. Number of natural and hatchery origin SPUN Chinook salmon (*Oncorhynchus tshawytscha*) that display three different life history strategies, 30-50mm estuary entry, 51-70mm estuary entry, and >70mm estuary entry. These sizes were selected from prior works that describe two different size classes in the out-migrating fry.

	30-50mm	51-70mm	>70mm	Total
Natural Origin	6	11	15	32
Hatchery Origin	1	25	42	68
Total	7	36	57	100

When observing the natural origin SPUN Chinook salmon strontium incorporation patterns, a distinct drop in strontium was detected during the freshwater stage of their life history, distinct from the drop associated with the maternal yolk sac (Fig. 10). The initial freshwater strontium concentration is around 500ppm, followed by a shift to around 200-300ppm before entering the estuary. This signal was absent from the hatchery component of the population, which instead remained at around 500ppm in terms of their strontium concentration after their exogenous feeding check. Our assumption here is that this indicates movement of natural origin Chinook salmon beyond the diversion dam and into the Tsolum confluence (Fig 11). On average, natural origin Puntledge summer Chinook move to the lower river at 42.8mm, ranging from 22.4 to 56.5mm (Fig 12). Around 18% of the natural origin Chinook entered the lower river above 50mm, while 78% entered the lower river between 30-50mm. The relationship between lower river entry size and estuary entry size was not significant ($p = 0.104$). Of the 25 fish that entered the lower river between 30-50mm, five entered the estuary at 30-50mm, ten entered the estuary

at 51-70mm, and 10 entered the estuary at above 70mm. Out of five individuals that entered at stream-type Chinook salmon size ranges, three entered the lower river in the 30-50mm size range. Follow-up investigations using the strontium isotope ratios can help to illuminate these patterns more clearly and validate this strontium drop is indicative of movement into the lower part of the river.

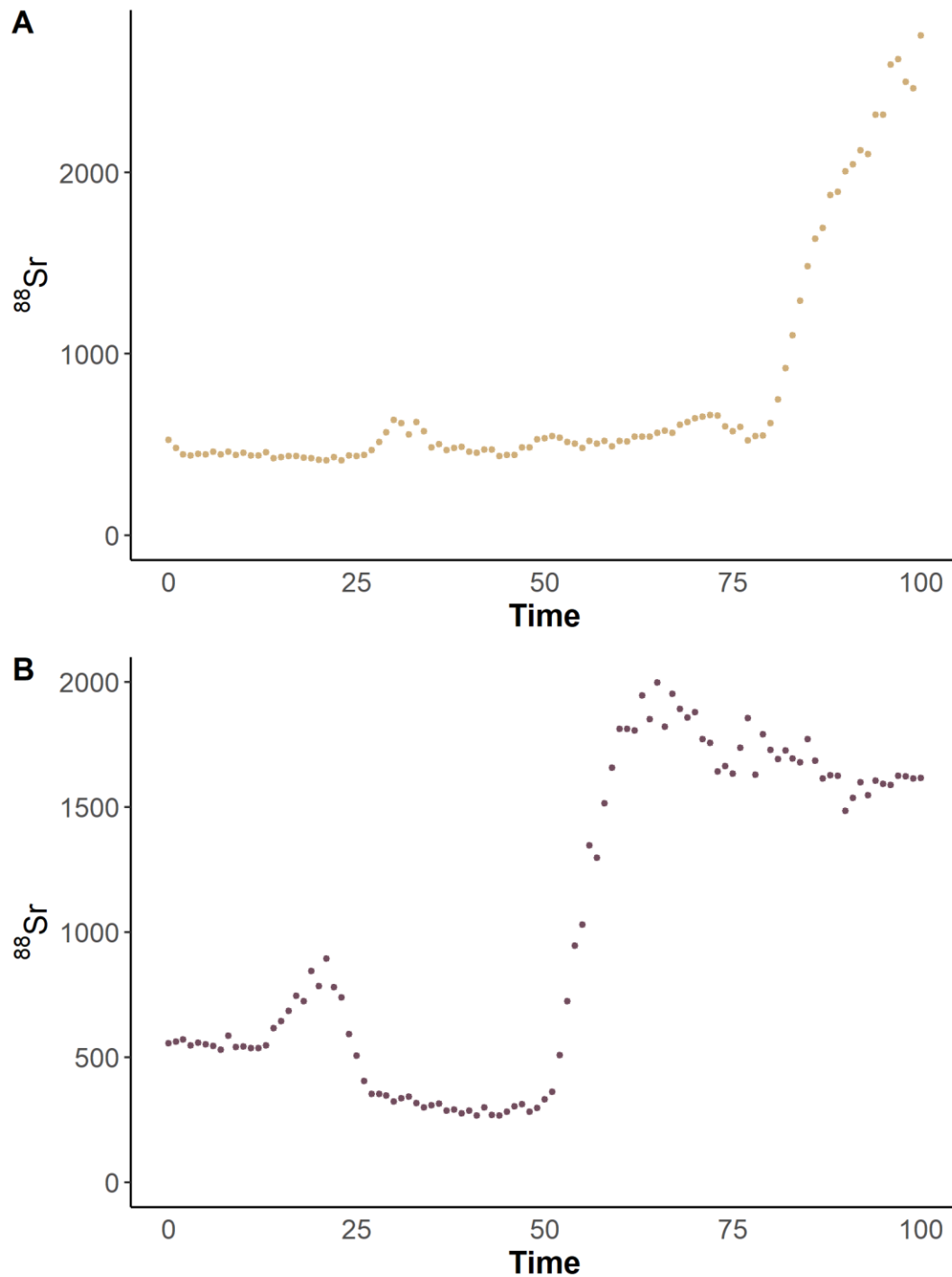


Figure 10. Strontium concentrations (ppm) over time (in seconds) for a hatchery origin (A) and natural origin (B) Chinook salmon (*Oncorhynchus tshawytscha*) from the summer run of the Puntledge River. The hatchery Chinook salmon have a more consistent strontium signal in the freshwater component of their life history (here between 0-75 seconds), while the natural origin Chinook salmon have two stages with a notable shift occurring around the 20 second mark. The rapid increase in strontium on the right side of both plots indicates movement into the estuarine environment.



Figure 11. Map of the lower Puntledge River system indicating the location of the Impoundment Dam at the start of the river (red star), the Diversion Dam (yellow star), the Puntledge River Hatchery (orange fish), the start of the Tsolum confluence (pink arrow), and the start of the estuary (green arrow).

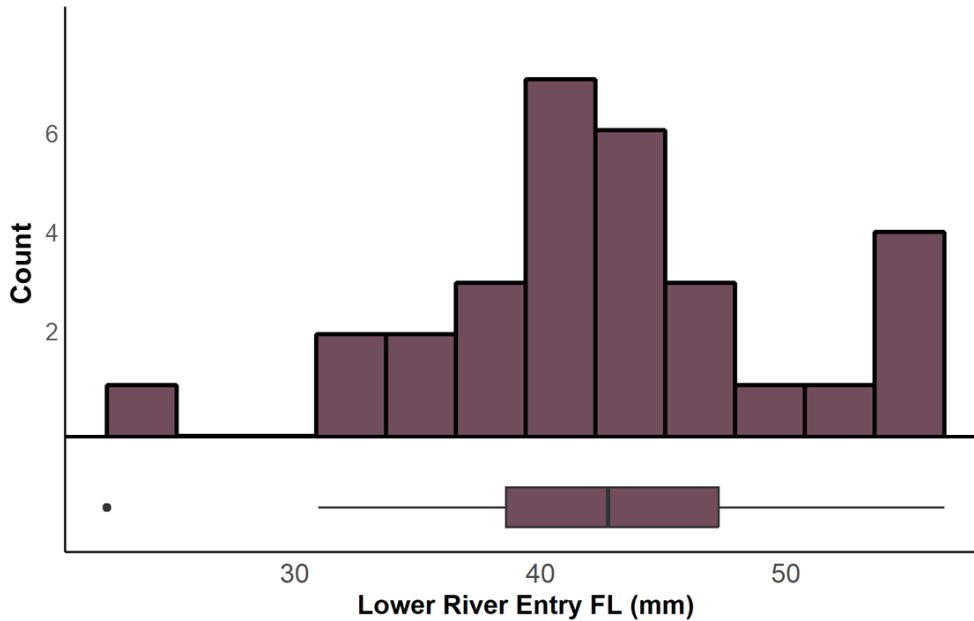


Figure 12. Histogram and boxplot of the lower river entry size of natural origin Chinook salmon. Mean size at lower river entry was 42.8mm. $n = 30$.

Nine of the ten Chinook salmon captured in the Puntledge River estuary were able to be analyzed for both estuary entry size and daily ring counts. The other individual was a hatchery origin Chinook salmon with two fully vateritic otoliths. These salmon were between 60 to 90mm. Size at estuary entry ranged from 36 to 84mm. Time spent in the estuary also ranged considerably, from as few as five days to 62 days. Estuary entry size was negatively correlated to the time spent within the estuary ($S = 212.780$, $p = 0.015$). Average growth rates also varied between these Chinook salmon, ranging from $3.35\mu\text{m/day}$ to $5.62\mu\text{m/day}$. Average growth rate was positively correlated with the size at estuary entry ($S = 25.606$, $p = 0.012$; Fig 13), though this is mainly due to the short distances between daily rings earlier on in the estuary as the last few rings were of similar distances across all Chinook salmon. The average growth rate in the estuary was $4.22\mu\text{m/day}$.

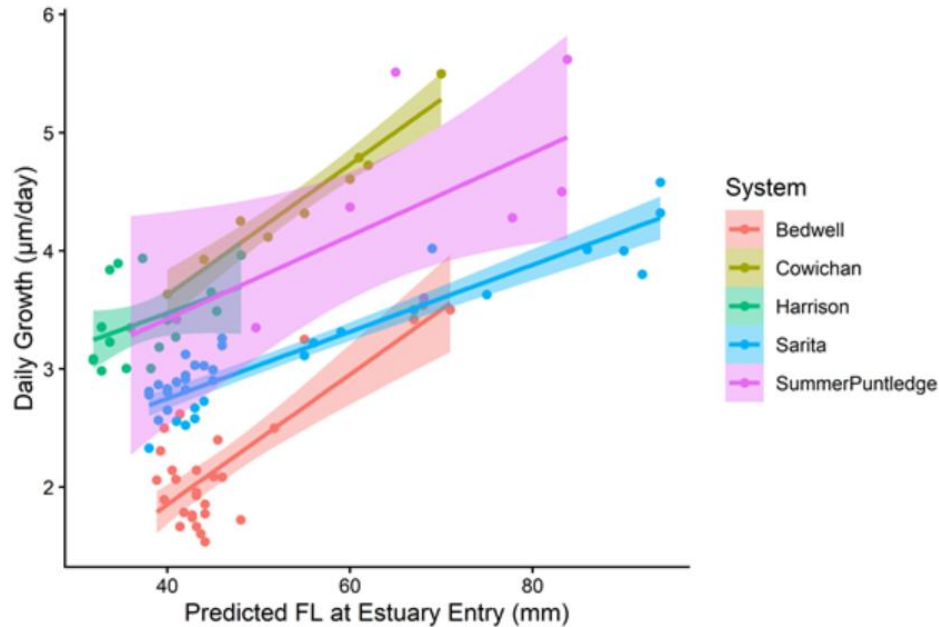


Figure 13. Scatterplot showing the relationship between the predicted size at estuary entry vs the number of days spent in the estuary for Chinook salmon (*Oncorhynchus tshawytscha*) smolts captured in various Southern British Columbia systems.

While the average size at estuary entry did not differ by origin, the time spent in the estuary did ($p < 0.001$; Fig 14). Natural origin Chinook used the estuary for far longer than hatchery origin Chinook (~21 days vs 9 days), and this difference was significant ($p < 0.001$). Eleven natural origin Chinook displayed extensive estuary use (more than 14 days) with one individual (SPUN21-68) spending a few months in the estuary. Larger outmigration life history types were less likely to use the estuary for prolonged periods of time (Table 4). Natural origin fry all utilize the estuary for extended rearing, while later and larger parr sized out-migrants do not use the estuary for long, instead moving out to other nearshore habitat in the Strait of Georgia.

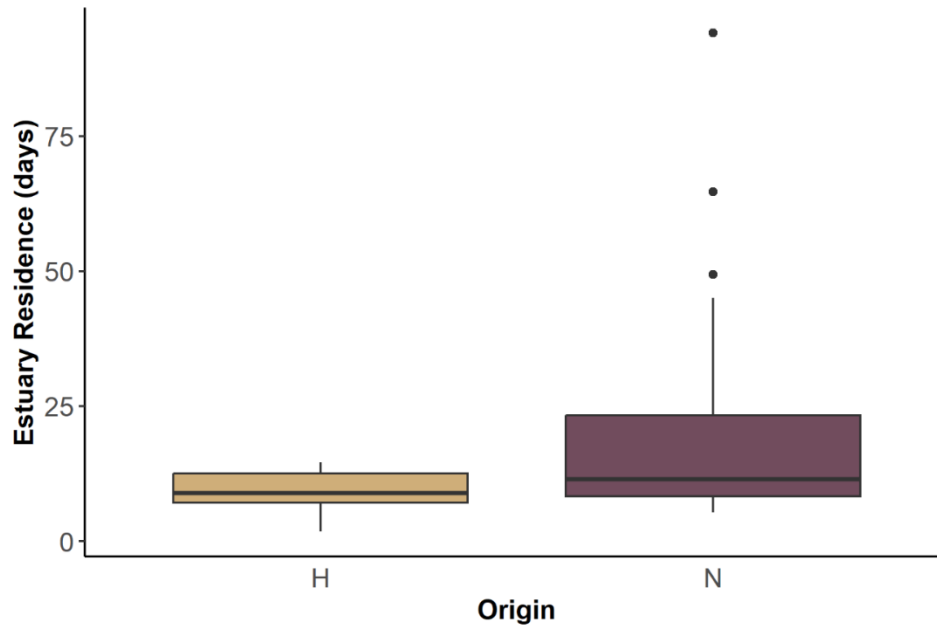


Figure 14. Predicted estuary usage in days for hatchery (gold) and natural origin (purple) summer Puntledge Chinook salmon (*Oncorhynchus tshawytscha*). n = 68 for hatchery origin fish and n = 32 for natural origin fish.

Table 4. Number of natural and hatchery origin Puntledge summer Chinook salmon that displayed extended estuary use (more than 14 days). Extended estuary usage is associated with using the estuary for more than transiting to the marine environment and conducting smoltification, i.e., they are rearing in this habitat. Natural and hatchery origin Puntledge summer Chinook are broken down into three life history categories based on predicted estuary entry size, 30-50mm, 51-70mm, and >70mm.

	Minimal Estuary Use	Extended Estuary Use	Total
30-50mm Natural Origin	0	6	6
30-50mm Hatchery Origin	0	1	1
51-70mm Natural Origin	6	5	11
51-70mm Hatchery Origin	25	0	25
>70mm Natural Origin	15	0	15
>70mm Hatchery Origin	42	0	42
Total	88	12	100

4. DISCUSSION

Otolith microchemistry provides fisheries scientists with the ability to analyze aspects of the life histories of salmon that are not obvious to the naked eye. Through LA-ICP-MS, the juvenile outmigration life histories of Puntledge summer Chinook were able to be identified in returning adult Chinook salmon.

4.1 SIZE AND AGE COMPOSITION OF HATCHERY AND NATURAL ORIGIN CHINOOK

Natural origin Chinook salmon were larger and older than their hatchery origin counterparts on average. While the jack rate of both groups was similar, 61% of the natural origin Chinook salmon were Gilbert-Rich age 4₁ while only 28% of hatchery origin Chinook were age 4₁. It should be noted that overall, females in both groups were typically four-year-olds (83%) and the major differences in age come from the male component of the population with 92.5% of all hatchery origin males being three years old or younger. There were some inconsistencies between scale and genetic based aging, particular amongst some of the oldest Chinook salmon in this study. These discrepancies seem to occur due to the late estuary entry of some of the older natural origin Chinook salmon, thus making them Gilbert-Rich age 5₂ rather than 4₁ as they were sometimes identified by scales. It is more likely that these scales were mis-aged due to an uncommon life-history strategy within the population. This highlights the utility of LA-ICP-MS analyses to uncover some of these uncommon life-histories within these populations. Most Vancouver Island Chinook salmon populations are pre-dominantly subyearlings (Teel et al., 2000), however, there are still yearling Chinook salmon that do occur in these populations. Subyearling and yearling Chinook salmon do not represent distinct genetic lineages, or at least not generally across all regions of the North Pacific but rather represent two distinct life-history strategies (subyearling and yearling outmigrants; Bourret et al., 2016). Growth rate often does affect the speed at which smolts emigrate from their river, with faster growing hatchery smolts often being predisposed to leave immediately upon release (Beckman et al., 1998). Our study supports this as hatchery individuals rarely spend any time in the estuary upon release but rather emigrate immediately into the ocean. This holds true for fall run Puntledge River Chinook salmon as well (Quindazzi, 2026). The presence of yearlings in the summer run Puntledge River population may be indicative of smaller, slower growing Chinook salmon that must emigrate in the following spring to compensate for slower early growth (Beckman et al., 2007). In order to assess the life history specific survival rates of outmigrating summer run Puntledge River Chinook salmon, further investigations of the composition of outmigrating smolts would need to be conducted.

4.2 VATERITE MEASUREMENTS

While not a priority within this study, vaterite prevalence and morphometric differences were also analyzed. Nearly every single otolith contained some calcite and vaterite, usually along the sulcus. This is consistent with previous findings (Wood et al., 2022). Extensively vateritic otoliths were found more within the hatchery origin SPUN Chinook with nearly half of these

Chinook having at least one predominantly vateritic otolith (Table 1). Only two of the 32 natural origin Chinook had at least one predominantly vateritic otolith. Age and sex appeared to have no effect on vaterite prevalence, which confirms previous findings (Quindazzi et al., 2023). While differences between hatchery and natural origin SPUN Chinook was most pronounced between 4-year-olds and amongst females, this is likely due to the larger number of 4-year-olds and females in the samples. Hatchery origin salmon typically have more vateritic otoliths than natural origin counterparts (Quindazzi et al., 2023; Gaffney et al., 2025) and this is likely due to increased growth rates or rearing densities (Austad et al., 2021; Reimer et al., 2017).

4.3 ESTUARY ENTRY AND RESIDENCY

Overall, the returning population appears to be dominated by juvenile life histories that do not make use of the estuary environment (88/100 Chinook). This is mainly due to considerable proportion of hatchery origin Chinook in this system, only 1% of which used the estuary for rearing (Table 4). In contrast, a sizeable proportion of the natural origin component did use the estuary for extended periods of time (34%). Of these 11 natural origin fish that use the estuary for a considerable amount of time, they spent an average of 43 days in the estuary. Of the 88 Chinook that did not use the estuary for more than two weeks, 28 remained in the estuary for less than seven days, which is within the margin of error of the incorporation delay of strontium into the otolith (Elsdon & Gillanders, 2002). This means it is very unlikely these fish spent time in the estuary outside of transiting through and conducting smoltification. These were mainly hatchery origin Chinook (21/28). The remaining 60 Chinook salmon did use the estuary for a short length of time (*i.e.*, between 1-2 weeks) before leaving. 14 of these Chinook were of natural origin, and 46 were hatchery origin. It is unclear if any of these fish did rear in the estuarine environment, or if these fish are simply transiting through and undergoing smoltification. The two week cutoff used here was based on experimental results which indicate that habitat transition between intermediate salinity environments is difficult to detect below a two week threshold (Miller, 2011).

While prolonged estuary usage occurred only in 34% of natural origin outmigrants, it was universal in the smallest 30-50mm outmigrant life history type (Table 4). While most of the hatchery origin Chinook entered the estuary above 50mm in size, these natural origin Chinook have multiple life histories in outmigrant size (Table 2). Each of these natural origin outmigration life history types has a different type of estuarine rearing pattern, while hatchery origin Chinook do not. It should be noted that one hatchery origin Chinook was released at quite a small size and also was the only hatchery fish which reared in the estuary (Table 4). Given the conservative estimate for what constituted estuary usage (more than 14 days), it is likely that the identified fish reflect those that rear in the system. As noted in other studies, subyearling Chinook rearing in the estuarine environment do so for weeks to over a month (Greene et al., 2025; Weitkamp et al., 2014). While there are no differences in average size at estuary entry between natural and hatchery origin fish (Fig. 9), there were differences in size at ocean entry (Fig. 15), which is driven by the extensive use of the estuary by components of the natural origin population (Fig 16). Clearly, some natural origin Chinook salmon in the Puntledge River do

make considerable use of the estuary. To further validate this inference, two individuals were captured as smolts having spent up to two months within the estuarine environment (Table 3). The Puntledge River estuary is smaller than the Fraser River estuary, but it appears to provide smolts there with similar scopes of growth (Chalifour et al., 2021). While the estuary may not provide enough habitat for all out-migrating smolts, it does provide good habitat for some proportion of the population, especially the small natural origin outmigrants.

Perhaps more interestingly, there was evidence that most natural origin Chinook (94%) use the lower river habitat below the Puntledge River Diversion Dam for considerable lengths of time. Interestingly, there was no relationship between size of individuals moving beyond the diversion dam to their subsequent estuary entry size for natural origin Chinook. In some cases, summer-run Puntledge Chinook appear to residualize in the river either leaving considerably late in the season, or the following year. While some of the fork lengths predicted were well outside our regression, they are still indicative of substantially large smolts in the natural origin population. The differences in habitat use also indicate that natural origin summer Puntledge Chinook are more at risk from the effects of historical logging, mining operations, human activities, runoff, higher temperatures, and predation. In particular, seal predation along two bridges in the lower river habitat, and in Comox Harbour is known to be quite high (Bigg et al., 1990; Yurk & Trites, 2000). Cover is uncommon in the lower part of the river and attempts to provide cover for Chinook smolts have failed in the past (Brown et al., 2003). Given the significant use of this habitat by natural origin summer run Puntledge Chinook salmon, efforts to conduct habitat restoration could provide smolts cover from high seal predation. The exact portion of the lower river used by these Chinook also needs to be identified to maximize the effectiveness of any restoration initiatives. It should be noted, however, that shifts in strontium incorporation in the otolith have been reported because of hatching, yolk sac absorption, and exogenous feeding (Chittaro et al., 2023), not habitat shifts. In this study, we also noted a change in strontium incorporation that appears to be an additional change to a new region with a different environmental signal. The most concrete evidence for this is the lack of such a signal in the hatchery Chinook salmon, and a near universal signal in natural origin Chinook salmon. Further investigations using the $^{87}\text{Sr}/^{86}\text{Sr}$ strontium isotope ratios would help to resolve this issue and identify movements within the freshwater environment more readily. Sentinel species such as members of *Cottus spp.* could also be used (Gray et al., 2018).

Natural origin summer run Puntledge Chinook appear to have more diverse life histories in terms of their estuary entry timing, estuary use, and return ages. While hatchery origin Chinook salmon can have broader genotypic diversity (Withler et al., 2018), natural origin Chinook have broader phenotypic diversity. Chinook salmon populations typically display risk reduction through a “bet-hedging” strategy to avoid population mortality (Healey, 1991; Stearns, 2000). Diversity in outmigration timing, estuary usage, and age at return are all examples of risk reduction in Chinook salmon (Hill et al., 2003; Schroeder et al., 2016). As seen in this study, early life history choices can have carry over effects into other decisions, such as the smallest Chinook outmigrants rearing extensively in the estuarine environment. Hatchery Chinook salmon, by contrast, typically have more variable survival rates once risk spreading via phenotypic diversity

is eliminated, which leaves populations at a greater risk of extinction during atypical years (Katz et al., 2013; Kilduff et al., 2019; Waples et al., 2009). The loss of phenotypic diversity in salmonid populations is an alarm bell for the loss of part if not all the population (Gustafson et al., 2007) and given that populations are not replaceable, the conservation of phenotypic diversity should therefore be a priority for the management of any salmon population. In the case of outmigration timing, if there is a significant mismatch between when the hatchery smolts are released and feeding conditions in the Strait of Georgia, marine survival could be significantly lowered. While most natural origin Chinook would also die, given they emigrate at the same time, the existence of other life histories ensure the population would survive this potential high stress event. Both components of the population therefore have critical roles to play in rebuilding a productive naturally spawning population.

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6. REFERENCES

- Austad, B., Vøllestad, L. A., & Foldvik, A. (2021). Frequency of vateritic otoliths and potential consequences for marine survival in hatchery-reared Atlantic salmon. *Journal of Fish Biology*, 98(5), 1401–1409. <https://doi.org/10.1111/jfb.14683>
- Beamish, R. J., Sweeting, R. M., Neville, C. M., Lange, K. L., Beacham, T. D., & Preikshot, D. (2012). Wild chinook salmon survive better than hatchery salmon in a period of poor production. *Environmental Biology of Fishes*, 94(1), 135–148.
- Beckman, B. R., Gadberry, B., Parkins, P., Cooper, K. A., & Arkush, K. D. (2007). State-dependent life history plasticity in Sacramento River winter-run chinook salmon (*Oncorhynchus tshawytscha*): Interactions among photoperiod and growth modulate smolting and early male maturation. *Canadian Journal of Fisheries and Aquatic Sciences*, 64(2), 256–271. <https://doi.org/10.1139/F07-001>
- Beckman, B. R., Larsen, D. A., Lee-Pawlak, B., & Dickhoff, W. W. (1998). Relation of Fish Size and Growth Rate to Migration of Spring Chinook Salmon Smolts. *North American Journal of Fisheries Management*, 18(3), 537–546. [https://doi.org/10.1577/1548-8675\(1998\)018<0537:rofsag>2.0.co;2](https://doi.org/10.1577/1548-8675(1998)018<0537:rofsag>2.0.co;2)
- Bigg, M. A., Ellis, G. M., Cottrell, P., & Milette, L. (1990). *Predation by harbour seals and sea lions on adult salmon in Comox Harbour and Cowichan Bay, British Columbia*.

- Bourret, S. L., Caudill, C. C., & Keefer, M. L. (2016). Diversity of juvenile Chinook salmon life history pathways. *Reviews in Fish Biology and Fisheries*, 26(3), 375–403. <https://doi.org/10.1007/s11160-016-9432-3>
- Brown, T. G., Munro, B., Beggs, C., Lochbaum, E., & Winchell, P. (2003). *Courtenay River Seal Fence*.
- Chalifour, L., Scott, D. C., Macduffee, M., Stark, S., Dower, J. F., Beacham, T. D., Martin, T. G., & Baum, J. K. (2021). Chinook salmon exhibit long-term rearing and early marine growth in the Fraser River, British Columbia, a large urban estuary. *Canadian Journal of Fisheries and Aquatic Sciences*, 78(5), 539–550. <https://doi.org/10.1139/cjfas-2020-0247>
- Chittaro, P., Hegg, J., Fuhrman, A., Robichaux, D., Doung, R., Beckman, B., Fisher, C., Vervoort, J., & Kennedy, B. (2023). The incorporation of environmentally derived $^{87}\text{Sr}/^{86}\text{Sr}$ and Sr/Ca in early otolith formation of Chinook salmon. *Ecology of Freshwater Fish*, 32(1), 107–119. <https://doi.org/10.1111/eff.12673>
- Cochran, W. G. (1954). Some Methods for Strengthening the Common χ^2 Tests. *Biometrics*, 10(4), 417–451.
- DFO. (1958). *The Fisheries Problems Associated with the Power Development of the Puntledge River, Vancouver Island B.C.*
- Elsdon, T. S., & Gillanders, B. M. (2002). Interactive effects of temperature and salinity on otolith chemistry: challenges for determining environmental histories of fish. *Canadian Journal of Fisheries and Aquatic Sciences*, 59(11), 1796–1808. <https://doi.org/10.1139/f02-154>
- Gaffney, L. P., Quindazzi, M., Polard, E., Kraemer, C., Walton, L. N., Molder, Z. A., Greentree, W., Duguid, W., Bohlender, N., & Juanes, F. (2025). Vateritic otoliths in hatchery-reared Strait of Georgia coho salmon: Variation among stocks, hatcheries, and life stages. *Fisheries Research*, 283, 107296.
- Gray, M. A., Allen Curry, R., Arciszewski, T. J., Munkittrick, K. R., & Brasfield, S. M. (2018). The biology and ecology of slimy sculpin: A recipe for effective environmental monitoring. *Facets*, 3(1), 103–127. <https://doi.org/10.1139/facets-2017-0069>
- Greene, C. M., Beamer, E. M., Munsch, S. H., Chamberlin, J. W., LeMoine, M. T., & Anderson, J. H. (2025). Population responses of Chinook salmon to two decades of restoration of estuary nursery habitat. *Frontiers in Marine Science*, 12, 1584913.
- Guimond, E., & Taylor, J. A. (2015). *Assessing emergent summer Chinook fry survival at the Puntledge diversion dam*.
- Guimond, E., & Taylor, J. A. (2017). *Evaluation of Natural and Hatchery Summer Chinook and Coho Production in the Upper Puntledge Watershed – 2016*.
- Guimond, E., & Withler, R. (2010). *Puntledge River Summer Chinook DNA Analyses 2009*.
- Gustafson, R. G., Waples, R. S., Myers, J. M., Weitkamp, L. A., Bryant, G. J., Johnson, O. W., & Hard, J. J. (2007). Pacific salmon extinctions: Quantifying lost and remaining diversity. *Conservation Biology*, 21(4), 1009–1020. <https://doi.org/10.1111/j.1523-1739.2007.00693.x>
- Healey, M. C. (1971). Utilization of the Nanaimo River Estuary by Juvenile Chinook Salmon, *Oncorhynchus tshawytscha*. *Fishery Bulletin*, 77, 653–668.

- Healey, M. C. (1991). Life history of chinook salmon (*Oncorhynchus tshawytscha*). In C. Groot & L. Margolis (Eds.), *Pacific salmon life histories* (pp. 311–394). UBC Press.
- Kalish, J. M. (1989). Otolith microchemistry: validation of the effects of physiology, age and environment on otolith composition. *Journal of Experimental Marine Biology and Ecology*, 132(3), 151–178. [https://doi.org/10.1016/0022-0981\(89\)90126-3](https://doi.org/10.1016/0022-0981(89)90126-3)
- Katz, J., Moyle, P. B., Quiñones, R. M., Israel, J., & Purdy, S. (2013). Impending extinction of salmon, steelhead, and trout (*Salmonidae*) in California. *Environmental Biology of Fishes*, 96(10–11), 1169–1186. <https://doi.org/10.1007/s10641-012-9974-8>
- Kilduff, D. P., Botsford, L. W., & Thompson, L. C. (2019). Frequency content of environmental variability and extinction risk of age-structured populations: Chinook salmon (*Oncorhynchus tshawytscha*) as an example. *Theoretical Ecology*, 12(2), 145–154. <https://doi.org/10.1007/s12080-018-0401-4>
- Lister, D. B. (1968). *Progress Report on Assessment of the Puntledge River Spawning Channel June 1965 to May 1968*.
- MacKinnon, C. N., Genoe, H., & Sinclair, D. C. (1979). *Puntledge River Project 1972-1977*.
- Mantel, N. (1963). Chi-Square Tests with One Degree of Freedom; Extensions of the Mantel-Haenszel Procedure. *Journal of the American Statistical Association*, 58(303), 690–700. <https://doi.org/10.1080/01621459.1963.10500879>
- Quindazzi, M. J., Gaffney, L. P., Polard, E., Bohlender, N., Duguid, W., & Juanes, F. (2023). Otolith mineralogy affects otolith shape asymmetry: a comparison of hatchery and natural origin Coho salmon (*Oncorhynchus kisutch*). *Journal of Fish Biology*, 102(4), 870–882. <https://doi.org/10.1111/jfb.15329>
- Quinn, T. P., Dickerson, B. R., & Vøllestad, L. A. (2005). Marine survival and distribution patterns of two Puget Sound hatchery populations of coho (*Oncorhynchus kisutch*) and chinook (*Oncorhynchus tshawytscha*) salmon. *Fisheries Research*, 76(2), 209–220. <https://doi.org/10.1016/j.fishres.2005.06.008>
- Reimer, T., Dempster, T., Wargelius, A., Fjellidal, P. G., Hansen, T., Glover, K. A., Solberg, M. F., & Swearer, S. E. (2017). Rapid growth causes abnormal vaterite formation in farmed fish otoliths. *Journal of Experimental Biology*, 220(16), 2965–2969. <https://doi.org/10.1242/jeb.148056>
- Stearns, S. C. (2000). Life history evolution: successes, limitations, and prospects. In *Naturwissenschaften* (Vol. 87). Springer-Verlag.
- Thomas, O. R. B., & Swearer, S. E. (2019). Otolith Biochemistry—A Review. *Reviews in Fisheries Science and Aquaculture*, 27(4), 458–489. <https://doi.org/10.1080/23308249.2019.1627285>
- Tripp, A., Morrison, S., Loeppky, A. R., & Davoren, G. K. (2022). Water strontium concentrations influence strontium concentrations in the pre-hatch otolith region of capelin (*Mallotus villosus*). *Journal of Experimental Marine Biology and Ecology*, 547. <https://doi.org/10.1016/j.jembe.2021.151667>
- Trites, A. W., Beggs, C. W., & Riddell, B. (1996). *Status Review of the Puntledge River Summer Chinook*.
- Waples, R. S., Beechie, T., & Pess, G. R. (2009). *Evolutionary History, Habitat Disturbance Regimes, and Anthropogenic Changes: What Do These Mean for Resilience of Pacific*

Salmon Populations? (Vol. 14, Issue 1).

<https://www.jstor.org/stable/26268053?seq=1&cid=pdf->

Weitkamp, L. A., Goulette, G., Hawkes, J., O'Malley, M., & Lipsky, C. (2014). Juvenile salmon in estuaries: comparisons between North American Atlantic and Pacific salmon populations. *Reviews in Fish Biology and Fisheries*, 24(3), 713-736.

Withler, R. E., Wetklo, M., Supernault, J., & Guimond, E. (2018). *Puntledge River Summer Chinook Parentage-based Tagging Study Year 4*.

Yurk, H., & Trites, A. W. (2000). Experimental Attempts to Reduce Predation by Harbor Seals on Out-Migrating Juvenile Salmonids. *Transactions of the American Fisheries Society*, 129(6), 1360–1366. [https://doi.org/10.1577/1548-8659\(2000\)129<1360:eatrpb>2.0.co;2](https://doi.org/10.1577/1548-8659(2000)129<1360:eatrpb>2.0.co;2)