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Early Life History and Stock Discrimination of Kokanee Salmon (Oncorhynchus nerka) in an Alpine Lake Environment

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EARLY LIFE HISTORY AND STOCK DISCRIMINATION OF KOKANEE SALMON (*ONCORHYNCHUS NERKA*) IN AN ALPINE LAKE ENVIRONMENT

A Thesis
Presented to
The Graduate Faculty
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In Partial Fulfillment
of the Requirements for the Degree
Master of Science
Biology

by
Alexandra Ruby McCarrel
November 2020

CENTRAL WASHINGTON UNIVERSITY

Graduate Studies

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ABSTRACT

EARLY LIFE HISTORY AND STOCK DISCRIMINATION OF KOKANEE SALMON (*ONCORHYNCHUS NERKA*) IN AN ALPINE LAKE ENVIRONMENT

by

Alexandra Ruby McCarrel

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This study examines an ecologically and recreationally important population of kokanee salmon (*Oncorhynchus nerka*) residing in Keechelus Lake and its tributary Gold Creek in the central Cascades of Washington State. This population of kokanee salmon is a vital food base for a population of ESA-listed resident bull trout. However, little is known about the early life history of this population and how it interacts with unique features in its rearing environment. With my research I described the early life history of kokanee salmon that spawn in the lake's main tributary, Gold Creek, and proposed a framework to determine the natal origin of spawning adults. Monitoring in the spring of 2019 showed spawning adults produced viable eggs that survived the winter with young-of-year emerging episodically in mid-April. The majority of adults avoided spawning in Gold Creek itself, preferring a man-made outlet channel that had significantly higher water temperatures and a prominent beaver dam. Stock discrimination of sampled spawning adults suggest that 83% of adults are of wild origin and 17% are of hatchery origin. The conservation of the Gold Creek Pond outlet channel will be critical for kokanee salmon spawning and rearing habitat until habitat becomes more suitable in Gold Creek. With this research, future restoration efforts in the Gold Creek ecosystem can integrate the life history data of kokanee salmon as well as assess the contribution of hatchery-origin kokanee salmon to the ecosystem.

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CHAPTER I

INTRODUCTION

This study examines a recreationally and economically important population of kokanee salmon (*Oncorhynchus nerka*) occupying Keechelus Lake and its tributary Gold Creek in the central Cascades of Washington State. Kokanee salmon in this population interact with unique features in the spawning environment, including a prominent beaver dam. The population was reestablished in the region for the purpose of recreational fishing through hatchery supplementation starting in the 1970s. Historically sockeye salmon runs, by access of the Yakima River on the south shore of Keechelus Lake, occupied the lake and Gold Creek (Deichl et al. 2011).

Sockeye Salmon

Sockeye salmon, *Oncorhynchus nerka*, is a Pacific salmon species ranging from Northwest Alaska to the Deschutes River in Oregon. Modern populations arose in the Pacific Northwest within the last 10,000 years at the end of the last ice age ensuing the retreat of the Cordilleran ice sheet (Lemay and Russello 2015). Sockeye salmon is one of the smaller sized species of Pacific salmon found in the Pacific Northwest, with an average length of 45-76 cm and an average weight of 1.8 to 6.8 kg (Burgner 1991). *O. nerka* is divided into four main ecotypes defined by their different life histories in fresh water (Quinn 2005; Lin et al. 2008; Whitlock et al. 2017; Beacham and Withler 2017). One ecotype is “lake-type” sockeye, typically spawning in lakes or tributaries adjacent to lakes with their offspring migrating to the ocean after one to three years rearing in said nursery lakes (Taylor et al. 1996). The lake-type sockeye is the most abundant and wide-spread of the sockeye ecotypes (Beacham and Withler 2017). Where lake habitats are unavailable, then the second ecotype, “river-type,” can be found. These sockeye salmon spawn in rivers or mainstem side channels, utilizing low-velocity sections of river as juveniles for one to two years before migrating to the ocean. Occasionally these juveniles migrate to the ocean as less than one year old after spending only a few months in their natal rivers. These ocean-rearing sockeye are known as “sea-type” sockeye salmon. Anadromous sockeye on average spend one to four years in the ocean before returning to spawn in their natal streams (Wood et al. 2008; Beacham and Withler 2017).

The fourth ecotype is termed “resident” sockeye, known as kokanee salmon, silver trout, or little redfish in North America. These non-anadromous salmon complete their entire life cycle in fresh water.

The most widely-accepted hypothesis of kokanee salmon evolution is that they evolved from ancestral “sea-type” sockeye salmon. This genetically diverse ancestral “sea-type” sockeye colonized new habitats after glacial retreat. “Lake-type” sockeye differentiated once lake habitat became accessible and productive, allowing for repeated adaptive radiations. Eventually the fitness of “lake-type” became equal to anadromous individuals and consequently populations of “resident-type” kokanee salmon evolved independently from “lake-type” sockeye (Taylor et al. 1996; Beacham and Withler 2017; Veale and Russello 2017). It is hypothesized that kokanee salmon are polyphyletic, arising from sockeye salmon on multiple independent occasions (Taylor et al. 1996). In today’s world, kokanee and sockeye salmon can co-occur in lakes and their tributaries where access to the sea is present, with hybridization between the two alternative life-history types possible (Beacham and Withler 2017).

Despite being evolved from common ancestors, physiological differences occur between kokanee and sockeye salmon. Kokanee salmon are typically smaller than anadromous sockeye salmon, averaging 22-30 cm in length and weighing 0.45 to 1.8 kg (Parametrix, Inc. 2003; Quinn 2005). Differences in size primarily arise from the reduced productivity of lakes compared to rivers (Parametrix, Inc. 2003). Markings and coloration of kokanee salmon are similar to anadromous sockeye, with bright silver sides, white bellies, and bluish-black tops before spawning. Spawning will bring coloration changes of bright red bodies with green heads with males developing characteristic humped backs and hooked jaws (Burgner 1991). Compared to anadromous sockeye salmon, kokanee salmon spawn earlier in the year, possess additional gill rakers, have poorer smolting ability, and tend to absorb scale margins at a greater degree upon maturity. Kokanee salmon on average sexually mature at four years of age and spawn and die in their natal freshwater streams (Quinn 2005). Occasionally they spawn along the shorelines of their nursery lake instead of spawning in freshwater streams (Beacham and Withler 2017). Strictly plankton feeders, kokanee salmon are known to prey on crustacean zooplankton, commonly known as water fleas, including *Daphnia*, *Holopedium*, *Bosmina*, and calanoid copepods (Hansen et al. 2017). Overall, kokanee salmon have filled a unique niche in the Pacific Northwest that multiple species depend on to survive.

Beaver Dam Effects on the Environment

The origin of this research project was a strong focus on the effects North American beavers (*Castor canadensis*) on kokanee salmon. Focus over the course of the study was eventually broadened beyond the impacts of beavers due to the unique ecological opportunity the study site had to offer.

Beaver activity has been shown to influence fish and wildlife populations, especially salmon. Chinook and coho salmon diversity can be impacted by beaver activity, with beaver dams on floodplain springs producing larger juvenile Chinook and coho salmon with increased total biomass (Pollock et al. 2011; Malison et al. 2015). Scientific literature lacks studies focusing on the effects beaver dams have on kokanee salmon specifically, with no scientific literature found on the subject when searched. With kokanee salmon being a popular sport fish in Washington State and listed as endangered in certain lakes in Washington (Lake Samammish), it is important to understand how beaver activity affects kokanee salmon populations.

Stock Discrimination

Pacific salmon are a vital part of life in the Pacific Northwest. With ecological, cultural, recreational, and economic importance, Pacific salmon species have a complex and well-researched past, especially due to their declining population numbers in the past century (National Research Council 1996). Pacific salmon, including Chinook, sockeye, coho, chum, and pink salmon, are estimated to have disappeared from approximately 40% of their historical ranges in the Pacific Northwest over the last century. Habitat degradation and destruction from forest clearing, agricultural, and urbanization practices, the construction of dams and other fish passage barriers, and commercial fishing all contribute to a shrinking salmon presence in the Pacific Northwest (National Research Council 1996). The Washington State salmon industry is worth millions of dollars, with money being spent on native fish recovery, ecosystem restoration, salmon stock production, and fishing licensing (Anderson and Larson n.d.). With salmon being such an important fixture in the Washington economy, any factor that could influence population health and distribution should be well analyzed. With such drastic population declines, drastic efforts have thus been taken to try and restore salmon populations.

Extensive revitalization efforts have been implemented for decades to try and increase salmonid populations to more stable levels. One result of this widespread effort is the massive investment in fish hatcheries. An estimated 80% of the fish found in Pacific Northwest salmon fisheries, approximately 325 million juvenile salmon in Oregon, Washington, and Idaho each year, come from hatcheries, with nearly 500 salmon hatchery programs in operation all over the region (Flagg 2015). Hatcheries have been used to try to artificially boost fish populations where fish stocks are threatened for more than 100 years (Brannon et al. 2004). Initially hailed as the saving grace to declining salmon populations, vast amounts of resources have been invested into developing and maintaining these hatchery facilities. In the beginning of fish hatchery implementation, hatchery fish were readily mixed in with threatened native salmon stocks. However, questions soon arose regarding the success of hatchery fish and how they contribute to wild salmon stocks. Were hatchery reared fish effectively recovering depleted or endangered salmon stocks or were they harming naturally spawned populations? With the addition of hatchery fish, the chances of over-harvest of wild fish when mixed with hatchery-origin fish are increased with the result in higher exploitation rates. This can in turn lower wild salmon population vitality to a greater extent (Brannon et al. 2004). The issues becomes even more complicated when hatchery fish start to interbreed with wild fish populations, creating the loss of genetic endemism of wild salmon populations (Kaeriyama and Edpalina 2004).

The life of a hatchery larva is one of luxury compared to its wild counterpart. Survival to fry rates greatly increase in hatchery salmon due to regular feeding, regulated water temperatures, and a round the clock crew to monitor fish health (Kaeriyama and Edpalina 2004; Brannon et al. 2004). Once hatchery fish are released into the wild, they have the potential to drastically alter the community interactions of their new home. Even if hatchery fish are made sterile, they still can outcompete native fish stocks through competition for habitat, food resources, and breeding ground (Brannon et al. 2004; Flagg 2015). Warmer water temperatures used in hatchery procedures can accelerate the onset of exogenous feeding, hasten maturation, and increase body size of hatchery salmon (Brannon et al. 2004). With such stark differences in early life history of native versus hatchery reared salmon, quantifying the extent hatchery salmon contribute to a given population is important to understand just how influential hatchery operations can be.

There are numerous benefits of developing a stock discrimination for a chosen fish population. Knowledge regarding the contribution of hatchery salmon to a native salmon stock can help track population trends of naturally spawned salmon independent of hatchery supplementation. Differences can be ascertained between naturally spawned and hatchery salmon on how the two groups respond to environmental variation and how they influence the environment in turn (Barnett-Johnson et al. 2007). For a management perspective, the ability to distinguish hatchery from wild-origin fish, while quantifying the survivors of each hatchery release cohort, can enable hatcheries to develop best stocking strategies (Paragamian et al. 1992). In order to determine natal origins of targeted fish populations, one low-cost method to do so is to analyze the variations in daily growth patterns and life history transitions of fish ear bones, called otoliths.

Otoliths are formed by the daily accumulation of calcium carbonate into a proteinaceous matrix. Exhibiting continuous growth, a pattern of “growth rings” form on the surface of the otolith chronologically, surrounding the central primordia (Brothers et al. 1976; Barnett-Johnson et al. 2007; Starrs et al. 2014). Each daily growth ring, also known as an increment, consists of a wide calcium-rich zone formed during daylight hours followed by a narrow calcium-poor incremental zone formed during the night (Geffen 1995). The buildup of rings can be inhibited by periods of physiological stress on the fish, such as water temperature changes, photoperiod changes, the hatching of a larva, and the transition to externally feeding for the first time. In turn these stressors leave discernible marks on the otolith, called checks (Paragamian et al. 1992; Barnett-Johnson et al. 2007; Freshwater et al. 2015). Hatchery-origin fish can lack a distinct exogenous feeding check formed from the abrupt transition of feeding exclusively on their maternal yolk sac to feeding externally for the first time (Barnett-Johnson et al. 2007). Because otoliths are formed chronologically, they have been shown to be dependable indicators of fish age. Experimental studies have shown that in periods of stress, there is no resorption of otoliths, which is not true for other structures that encode age information such as fish scales and vertebrae. Thus otoliths, once properly prepared, show a permanent record of life history events (Jones 1992; Geffen 1995).

Otoliths can be used as “natural” tags to differentiate between hatchery-origin and wild-origin fish stocks as long as they experience different early life history growth environments (Zhang et al. 1995; Barnett-Johnson et al. 2007). Kokanee salmon exhibit daily otolith increments with discernible otolith “stress” checks that can be used to compare different early life history rearing environments (Paragamian et al. 1992). Past scientific literature has shown that it is possible to conduct a stock discrimination using otolith microstructure analysis. A study by Zhang et al. (1995) researched specific otolith microstructure differences between hatchery-reared and wild-reared Chinook salmon from the Cowichan River in British Columbia. Using a combination of larvae, smolts, and adults, stressor checks and the pattern of daily growth increments were analyzed. Daily growth increments that developed immediately after the exogenous feeding check were more uniform in width for hatchery-reared Chinook compared to wild-reared Chinook salmon. In addition, hatchery-reared Chinook also possessed the presence of a check formed when the fish were released from the hatchery. The authors were successfully able to identify rearing origins of 89% of a sample of 67 Chinook smolts using these parameters (Zhang et al. 1995).

A study by Barnett-Johnson et al. in 2007 also looked at the stock discrimination of hatchery and wild Chinook salmon using otolith microstructure analysis. Supporting the finding of Zhang et al., the authors established a framework to distinguish the rearing origins of Chinook salmon by analyzing the exogenous feeding check. The daily growth increments (~30) immediately after the exogenous feeding check were wider and more uniform in width compared to those of wild-rearing-origin fish. The authors had a 91% success rate in identifying the natal origins of Chinook salmon across years, life history stages, and geographic regions (Barnett-Johnson et al. 2007). Using the methods and parameters developed by Zhang et al. and Barnett-Johnson et al., a stock discrimination is critical in quantifying the contribution hatchery-reared salmon have on their local ecosystem, especially in cases where hatchery-reared salmon are not physically marked.

Objectives

This study had two main research goals. The first goal was to evaluate the early life of history kokanee salmon that reside in the Gold Creek ecosystem, a headwater third-order stream located east of Snoqualmie Pass, and their interactions with unique habitat features. The first question was, 1) Do kokanee salmon that spawn in the Gold Creek ecosystem produce viable offspring that survive the winter and emerge in the spring and if so, how do these young-of-year larvae interact with unique features in their rearing environment (an established beaver dam and the presence of two

Interstate 90 bridges)? The second part of the study was to conduct a stock discrimination of spawning adult kokanee salmon in the Gold Creek ecosystem. The second question was, 2) What are the natal origins (wild-reared versus hatchery-reared) of adult kokanee salmon that spawn in the Gold Creek ecosystem?

CHAPTER II

STUDY AREA

Gold Creek, a headwater, third-order stream, originates in the Alpine Lakes Wilderness Area and flows south-southwest into Keechelus Lake (Figure 1). Gold Creek flows underneath Interstate 90 on the eastern side of Snoqualmie Pass in the Cascade Mountain Range, Kittitas County. The stream flows for approximately 13 km before entering the north end of Keechelus Lake, a natural lake turned into an irrigation reservoir in 1917. Gold Creek is located within the Yakima River drainage with a catchment size of 36 km^2 and a mean velocity of 2.61 m/s (Wissmar and Craig 2004; Deichl et al. 2011; Kittitas Conservation Trust 2019).

Physical Habitat

Gold Creek catchment has a maximum elevation of 1597 m above mean sea level (AMSL) and a minimum elevation of 762 m AMSL where it enters Keechelus Lake. Gold Creek flows within a u-shaped glacier-carved valley, with Rampart Ridge bordering to the southeast and Snoqualmie Pass and Kendall Peak to the northwest. The climate of the area is cool and wet with an average annual precipitation at Snoqualmie Pass of 213.74 cm. Temperatures in the area fluctuate between -6 °C to 10 °C on average between winter and summer months (Deichl et al. 2011).

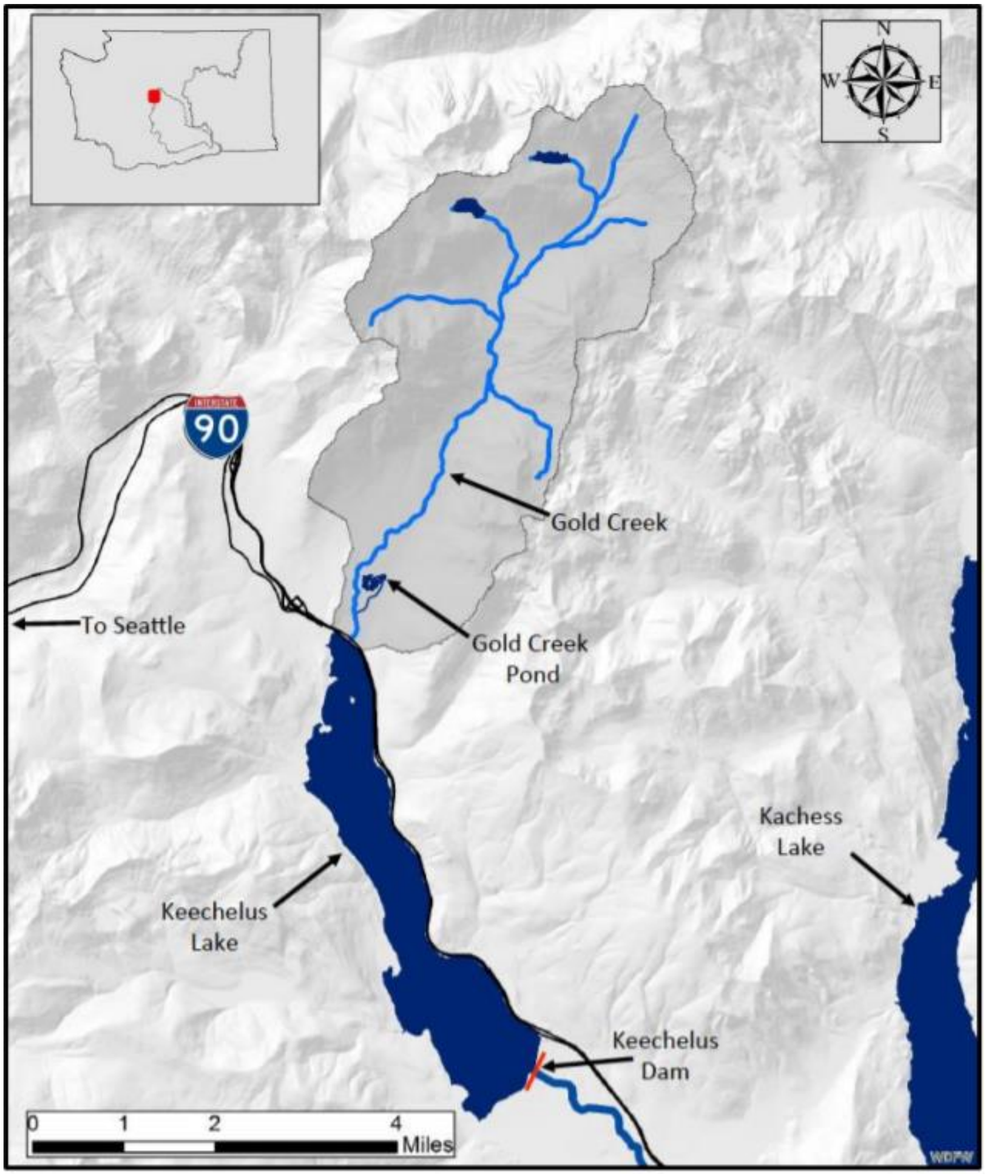

Figure 1. Image showing the Gold Creek watershed and surrounding environment near Snoqualmie Pass, Washington.

The lower reach of Gold Creek (river mile 0-1.85) is a low-gradient (1%) braided channel flowing over highly permeable gravel and sand. It has an average bankfull width of 49 m and a valley width of 152 m. The more confined middle reach (RM 1.85-3.1) has a bankfull width of 15 m and a valley width of 30.5 m with an average gradient of 3%. The middle reach is composed mainly of pool-riffle segments with cobbles and boulders.

The upper reach of Gold Creek (RM 3.1-7.1) is moderately confined. It has a bankfull width of 12 m, a valley width of 30.5 m, and is high-gradient (5%), consisting of step-pool segments underlain by bedrock and boulders. The lower reach of Gold Creek has a stream discharge ranging from 0.35 m^3/s in mid-August to 0.56 m^3/s in late September (Natural Systems Design 2013).

The geology of Gold Creek is complex. Bedrock consists of highly folded and faulted rhyolitic and basaltic volcanic rocks, with the formation having an approximate thickness between 1524 and 3048 m. Surficial geology includes dense glacial drift and outwash sediments with higher percentages of gravel. The near-surface geology of Gold Creek is variable, with the area east of Gold Creek consisting of Eocene and Miocene-age volcanic and igneous rock. To the west of Gold Creek near-surface geology consists of primarily younger unconsolidated sediments. The drainage basin itself is primarily made up of alluvial deposits. During the construction of Interstate 90 (I-90), surveys showed numerous major sedimentary units and one bedrock unit beneath Gold Creek Valley. The topmost unit consists of sand, cobbles, and boulders ranging 2.7 to 6.4 m thick. Beneath this layer is a layer of organic soil, ranging 0.3 to 4.6 m thick. A layer of native soil, consisting of gravel with sand and cobbles, underlies the organic soil layer, with a thickness of 1.8 to 7.3 m. Finally, fine sands and silt underlies the native soil (Deichl et al. 2011).

Biological Habitat

Prior to the construction of the Keechelus Lake dam in 1917, anadromous sockeye salmon migrated up the Yakima River and spawned in Gold Creek. The construction of the dam raised water levels and created a fish passage barrier. Chinook salmon and summer steelhead occupied the Keechelus Lake region up until the 1917 dam construction (Deichl et al. 2011; Hansen et al. 2017). It is unknown if resident ecotype kokanee salmon resided in Keechelus Lake alongside anadromous sockeye salmon prior to the dam's construction. After the construction of the dam, sockeye salmon in the form of kokanee salmon were reintroduced into the system through hatchery supplementation starting in the 1970s (Deichl et al. 2011). Kokanee salmon today spawn in large numbers in Gold Creek and the Gold Creek Pond outlet channel. Gold Creek and its channels are the only location kokanee spawn in, ignoring all other tributaries flowing into Keechelus Lake. Kokanee salmon are stocked in Keechelus Lake by the Washington Department of Fish and Wildlife (WDFW) and are popular for recreational fishing (WDFW n.d.).

Historically bull trout, *Salvelinus confluentus*, could be found in major tributaries of the Columbia River on the eastside of the Cascades in Washington State. Bull trout were listed as "threatened" by the federal Endangered Species Act (ESA) in November 1999 and critical habitat was designated in 2005. Bull trout most likely existed as isolated adfluvial stock in Keechelus Lake and spawned in Gold Creek pre and post-dam construction (Hansen et al. 2017). Populations, however, are declining, with an estimated population of less than 50 adults still existing today (Hansen et al. 2017). Reasons for Gold Creek bull trout population declines include the habitat alterations from the Keechelus Lake dam construction, impacts from nearby logging, road development, urbanization, and the seasonal dewatering of Gold Creek (U.S. Fish and Wildlife Service 2002; Deichl et al. 2011; Mizell and Anderson 2015).

A unique aspect of this study is that bull trout in Keechelus Lake depend on kokanee salmon, plus other pelagic fish such as pygmy whitefish (*Prosopium coulterii*) and redside shiner (*Richardsonius balteatus*), for the majority of their diet (Hansen et al. 2017). Approximately 300,000 kokanee salmon fry are stocked into Keechelus Lake mid-summer each year. Within the past decade, the hatchery that stocks fry into Keechelus Lake, the Naches Fish Hatchery run by WDFW, has faced multiple instances of budget cuts that put in jeopardy future fry stockings. (Thompson 2019). With bull trout numbers dangerously low, any significant changes to Keechelus Lake kokanee salmon stocking has the potential to negatively impact the vulnerable bull trout population by jeopardizing a potentially important food source.

Site History

The region of Gold Creek is estimated to have been inhabited by the Kittitas Native Americans, part of the Shahaptian tribal group and of the coastal Native Americans west of the Cascades. Tribal people were thought to use Gold Creek and the Keechelus Lake areas for berry foraging, fishing, and game hunting. It is believed that coastal and plateau Native Americans likely established a summer campsite at the mouth of Gold Creek at its connection point to Keechelus Lake. A total of 58 cultural resources were identified in the Washington State Department of Transportation (WSDOT) I-90 Snoqualmie Pass East Project area, which includes the region of Gold Creek and Keechelus Lake. The locations of these sites are undisclosed to preserve the integrity of the sites (Deichl et al. 2011). Contact with European settlers began in the 1800s, with logging, mining, and railroad enterprises beginning in the mid-late 1800s.

As human populations increased due to an influx of pioneers, improvements to transportation infrastructures were made. A well-established Native American commerce trail was used by pioneers to cross Snoqualmie Pass starting in the 1850s until the Snoqualmie wagon road was built. The Chicago, Milwaukee and St. Paul Railroad (formally the North Pacific Railroad Company) was completed over Snoqualmie Pass in 1909 (Deichl et al. 2011).

In 1880 mining operations began in the Gold Creek area. Coal mining, as well as precious metals such as gold, silver, copper, lead, and many others were placer mined in the region (Figure 2). At least three major mining companies operated along Gold Creek during the end of the 19th century; Esther, Giant, and Granite King Mines. Numerous smaller mining claims existed in the region as well, with mining activity peaking in the 1930s (Deichl et al. 2011; Natural Systems Design 2013).

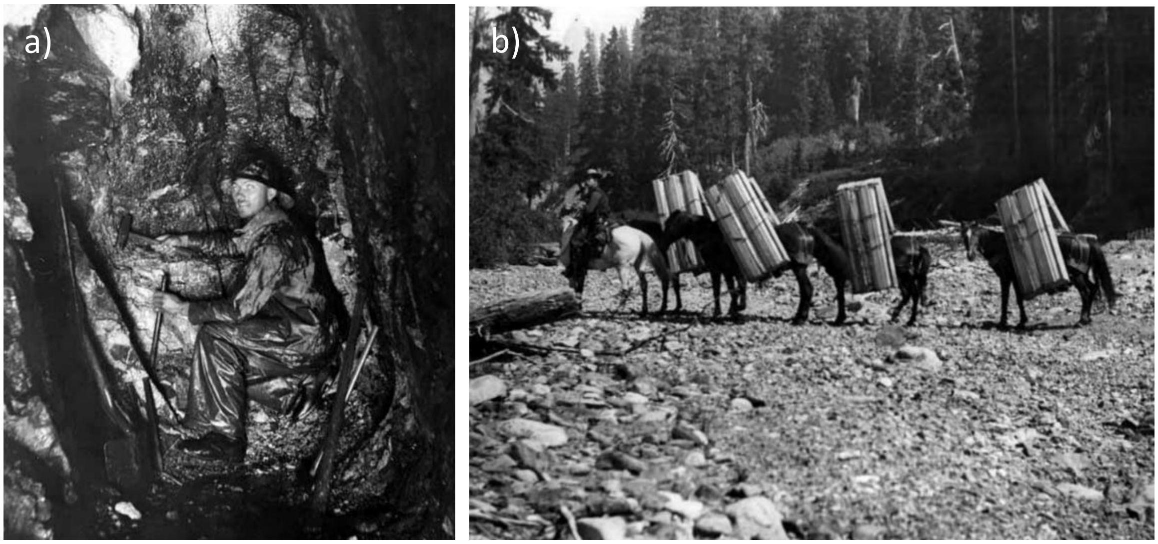

Figure 2. Photograph of a) miner Lewis Witt in a tunnel inside Esther Mine, near Gold Creek, 1898, and b) pack train of horses led by Tom Denny carrying ventilation pipes used for mining, Gold Creek, 1898, UW Libraries, Lawrence D. Lindsley Photographs Collection.

Along with mining, the timber industry of Gold Creek was strong. Timber removal was estimated to have begun during the construction of a mining tramway into Gold Creek valley in the late 1800s (Natural Systems Design 2013). In 1902 the watersheds on the eastern slopes of the Cascades, including Gold Creek, were evaluated for watershed and timber harvest health. Reports shows that the Cascade forests were considered free of major diseases or defects during the beginning of the 1900s. Early forest fires were documented in some areas surrounding Gold Creek, resulting in second growth forests. In 1941 merchantable fir trees were harvested and a timber survey for Kittitas County was conducted between 1956 and 1959 in Gold Creek and Keechelus Lake. The North Pacific Railroad Company owned the land in a checkerboard pattern along with the U.S. Government and gradually sold the land to the United States Forest

Service, United States Bureau of Land Management, and various private timber companies in the mid-19th century.

Approximately 1.6 km upstream of the confluence of Gold Creek and Keechelus Lake is Gold Creek Pond. Originally a wetlands habitat complex, the area was excavated to provide an expansive gravel pit, dug by the Washington Department of Transportation in the 1970s to help with the construction of the Sunset Highway, I-90, and Forest Service Road #4832 (Deichl et al. 2011). The location of I-90 follows the same path as the historical Snoqualmie wagon road used by pioneers in the 1850s. The gravel pit (PS-S-156) is estimated to take up 90% of the floodplain with a maximum depth of 18.3 m. It is one of the few gravel pits unrestored to its original state by WSDOT after construction (Washington State Recreation and Conservation Office 2018; Kittitas Conservation Trust 2019). The pit site was estimated to be composed of mostly gravel backfill, mineral aggregate for asphalt concrete, sand, cement concrete aggregate, ballast, and crushed surfacing with an estimated haul of 42,245 m^3 (Deichl et al. 2011).

Starting in 1970, gravel spawning beds were constructed for salmonids on the site of the PS-S-156 gravel pit. The gravel spawning beds flow for 0.4 km until it reaches Gold Creek. An extension of the PS-S-156 gravel pit was constructed next to a naturally existing pond, creating the groundwater fed, oligotrophic Gold Creek Pond we see today. In addition, WSDOT constructed a pervious dam between the outlet channel and Gold Creek Pond. The construction of Gold Creek Pond confined Gold Creek to the western most boundary of its historical floodplain (Deichl et al. 2011).

Due to the steep nature of the gravel pit and the elevation of its downstream outlet channel, the groundwater table lowered across Gold Creek Valley. The site of the former gravel pit acts as a siphon, drawing water away from Gold Creek. This is turn contributes to severe seasonal dewatering of Gold Creek (Kittitas Conservation Trust 2019). The construction of Keechelus Lake dam in 1917 originally led to reduced water levels in Gold Creek, creating a barrier to fish migration (Natural Systems Design 2013). The original lakeshore moved northward ~2 km with the water level rising upwards of 18.6 m, changing the drainage basin of Gold Creek by inundating the basin during spring high lake levels (Deichl et al. 2011).

In 1978, WSDOT widened I-90 over Gold Creek and incorporated restoration efforts of re-vegetation surrounding PS-S-156. Vegetation planted in the Gold Creek floodplain in the 1970s included 1,750 Douglas fir (*Pseudotsuga mensiesii*), 875 grand fir (*Abies grandis*), 875 lodgepole pine (*Pinus contorta latifolia*), 15,791 big whortleberry (*Vaccinium membranaceum*), 720 mountain ash (*Sorbus americana*), 19,125 creeping snowberry (*Symphoricarpos orbiculatus*) , and 488 vine maple (*Acer circinatum*). The Keechelus-Kachess Subbasin watersheds themselves are dominated by western hemlock (*Tsuga heterophylla*) and Pacific silver fir (*Abies amabilis*) trees with subalpine species present at higher elevations (Deichl et al. 2011). In 2013, WSDOT replaced the previous constrictive 42.7 m bridge over Gold Creek with two new bridges approximately 274.3 and 335.3 m in length. The construction allows for wildlife passage during high water levels of Keechelus Lake (Deichl et al. 2011). However, Gold Creek is still constricted by the presence of Forest Service Road #4832 roughly 50 m upstream of the I-90 transportation bridges.

CHAPTER III

METHODS

Study Site

The study site is the lower reach of Gold Creek (47° 24’19.1’’N, 121°22’32.0’’W), just east of Snoqualmie Pass in the Cascade Mountain Range, Washington. More specifically, research took place in the Gold Creek main channel and a distinct side outlet channel that runs from Gold Creek Pond to the Gold Creek main channel approximately 0.4 km downstream. The study site also included Gold Creek directly underneath the I-90 undercrossing just south of Forest Service Road #4832.

Larvae Capture

Kokanee salmon larvae were collected using a combination of submerged emergence traps and drift nets during spring 2019. A row of two drift nets (Figure 3) was placed one m south of the eastbound I-90 bridge in Gold Creek, inserted into the substrate using iron stakes. The collection bottle at the end of each drift net was oriented downstream with the drift net tops inserted above the water line to prevent larvae from passing above the net. At 0.4 km upstream of the I-90 undercrossing a row of two drift nets was placed five m below the beaver dam and another row of two drift nets placed 59 m above the beaver dam. Drift net placement was determined by the closest viable placement of nets in regards to stream structure and velocity (Figure 3). A row of two drift nets was placed in Gold Creek located 21 m above the confluence of Gold Creek and the Gold Creek Pond outlet channel. A third drift net was added to Gold Creek on May 1st, May 4th, and from May 15th to May 24th, 2019. Twelve larvae capture events were conducted between April 28th, 2019 and May 29th, 2019. The exact time of each drift net deployment was recorded. GPS coordinates for each drift net and fry emergence net location can be seen in Table 1.

Figure 3. Photographs of drift nets a) downstream of the beaver dam in the Gold Creek Pond outlet channel b) upstream of the beaver dam in the outlet channel c) Gold Creek and d) Gold Creek downstream of the I-90 bridges.

Table 1. GPS coordinates for fry emergence and drift nets in Gold Creek and the Gold Creek Pond outlet channel.

Net	Latitude	Longitude
Emergence Net 1	47°23'48.3"N	121°22'52.0"W
Emergence Net 2	47°23'51.4"N	121°22'51.2"W
Emergence Net 3	47°23'52.6"N	121°22'45.0"W
Above beaver dam drift net row	47°23'52.6"N	121°22'51.0"W
Below beaver dam drift net row	47°23'50.1"N	121°22'51.6"W
Gold Creek drift net row	47°23'45.8"N	121°22'57.4"W
Gold Creek by I-90 bridges drift net row	47°23'24.5"N	121°22'59.8"W

Three fry emergence nets were installed in the Gold Creek Pond outlet channel on April 28th, 2019. Locations of fry emergence trap sites were determined from spawning surveys conducted in October 2019. Sites of kokanee salmon redds were marked with flagging and GPS coordinates taken in order to maximize chances of fry emergence nets being placed over specific redds. For each emergence trap, a trench was dug by hand due to the excess of cobbles and boulders with the edges of the net buried underneath rocks and gravel to create a tight seal. Emerged larvae were captured in collection bottles located at the downstream end of the emergence traps (Figure 4). One fry emergence trap, Enet1, was placed 114 m downstream of the beaver dam, a second (Enet2) installed 38 m upstream of the beaver dam, and a third (Enet3) installed 85 m above the Gold Creek Pond outlet channel footbridge.

Figure 4. Photographs of fry emergence nets a) emergence net 1 (Enet1) b) emergence net 2 (Enet2) and c) emergence net 3 (Enet3).

Fry emergence traps were left in the field once installed with collection bottle caps removed in between capture events to prevent larvae becoming trapped between collection days. Collection bottle caps were installed during capture events with the exact time of installation and removal being marked. Each drift net and fry emergence trap was left collecting for a minimum of one hour before checking. Collecting was done during dusk hours as that is when fish are known to be the most active.

Drift nets and fry emergence traps were checked for larvae starting with the location first installed. Typically drift nets were first installed underneath the I-90 bridges, then at the site below the beaver dam and above the beaver dam in the Gold Creek Pond outlet channel, and finally in Gold Creek itself. The locations of each drift net row were marked with flagging tape to minimize differences in collection locations (Figure 5).

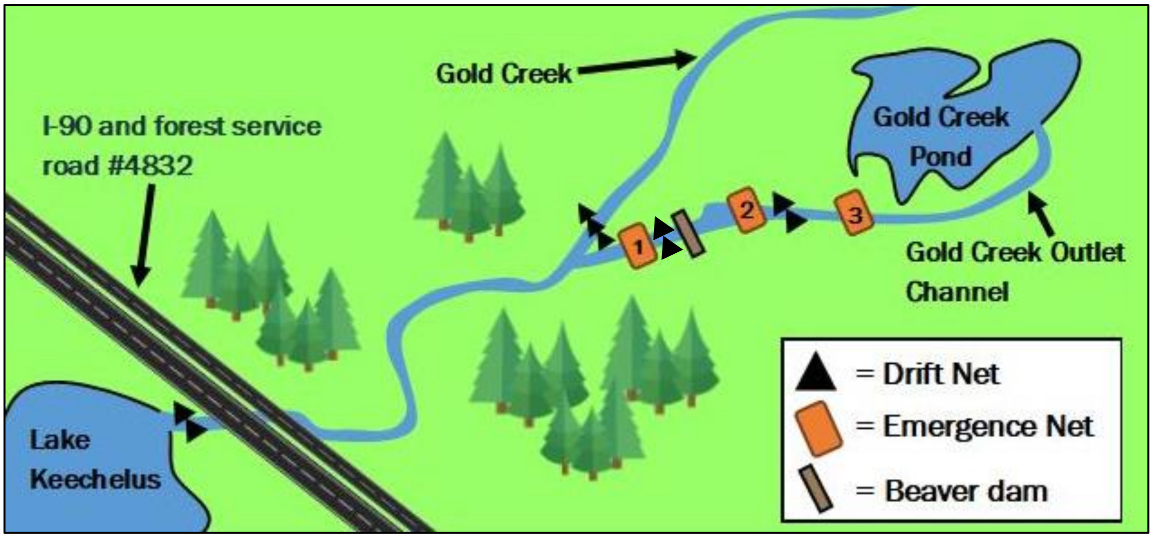

Figure 5. Site map of drift net sample locations for kokanee salmon larvae in Gold Creek and the Gold Creek Pond outlet channel.

Larvae Assessment

Once collected, larvae were placed in a five gallon bucket filled with fresh creek water and with an oxygenator. A solution of Tricaine Methanosulfonate (MS-222) was used to sedate fish for handling and measuring. Once the fish were adequately sedated (determined as the point at which the fish began to lose equilibrium balance, but with continuous gill movement) they were placed on a measuring board. Total length and width for each larva was recorded. Larvae then were analyzed to determine their developmental level. Methods were based on a similar study by Fuhrman et al. (2017), which divided the development of Chinook salmon into five distinct stages (Figure 6). Kokanee salmon larvae were visually inspected for development levels and given a score of 1-5 depending on the amount of yolk sac remaining with 1 being the most underdeveloped and the highest amount of yolk sac remaining and 5 being no visible yolk sac remaining.

Figure 6. Chinook salmon developmental stages scale (1-5) corresponding to amount of yolk left at emergence (Fuhrman et al. 2017).

An additional subsample was taken from each sample and euthanized by a lethal dose of MS-222, at a minimum dose of 250 mg/l recommended by the 2013 American Veterinarian Medical Association Guidelines for the Euthanasia of Animals. Each subsample averaged one to two larvae per collection day location due to the low numbers of larvae collected during each event. The euthanized larvae were then preserved in 70% ethanol and returned to the Central Washington University aquatic lab for later otolith extraction. The remaining fish, once measured and analyzed, were put into a recovery five gallon bucket with an oxygenator attached. Larvae were deemed suitable for release once they were upright and swimming continuously for at least a full minute. Larvae were released within the same 10 m of stream they were collected from. Development levels and body measurements were compared in order to gain an accurate understanding of the size, development, and maturity of the newly emerged salmon and any potential differences in the presence of the beaver dam or the I-90 bridges.

Auxiliary Habitat Measurements

Water velocity was measured at each drift net opening and recorded using a water velocity flow meter attached to a top-setting wading rod. The number of larvae per 100 m^3 was calculated for each drift net using the velocity of water for each drift net and the total time in seconds each drift net was actively collecting larvae.

To monitor water temperature, I installed six Onset® HOBO thermographs at my study site on October 27th, 2018. They were programmed to log the surrounding water temperature every four hours. One thermographs was placed underneath the I-90 bridges at the southernmost edge, one five m below the beaver dam and one five m above the beaver dam in the Gold Creek Pond outlet channel, one at the mouth of Gold Creek Pond in the Gold Creek Pond outlet channel, and two in Gold Creek 30 m apart. Water temperature data was collected in Gold Creek until 3/18/19 when I was forced to remove the devices due to high velocity of Gold Creek with heavy spring rain. Groundwater well measurements were taken by Natural Systems Design in 2019, with

Gold Creek groundwater being used as a proxy for Gold Creek Pond outlet channel water temperature measurements because of the close proximity of the outlet channel to the groundwater well and the fact that the outlet channel is fed by Gold Creek groundwater (Kittitas Conservation Trust 2019). Water temperatures taken during May 2019 in the Gold Creek Pond outlet channel were unavailable for this study.

Ambient maximum and minimum air temperatures were sourced from the Northwest Avalanche Center's Snoqualmie Pass weather station 3.7 km NW of Gold Creek (47°25'29.6"N 121°24'50.3"W, 917 m elevation).

Carcass Collection for Stock Discrimination

Kokanee salmon spawning carcasses were collected from Gold Creek on October 11th, 2019 and October 18th, 2019 towards the beginning of their spawning period (Figure 7). A second sample of carcasses was collected on November 15th, 2019 near the end of the spawning period. Carcasses were collected from Gold Creek and the Gold Creek Pond outlet channel and combined, in order to avoid sample bias. Once collected, kokanee salmon were transported back to the Central Washington University aquatic lab and frozen until otolith extraction. Fish were allowed to thaw approximately 12 hours before otolith extraction.

Figure 7. Kokanee salmon spawning downstream of the beaver dam in the Gold Creek Pond outlet channel, taken at the beginning of their spawning period on October 18th, 2019. Note the dam beaver dam being constructed downstream of the spawning adults.

Otolith Removal and Processing

Sagittal otoliths of the euthanized Gold Creek larvae collected in the spring of 2019 were removed using a dissection microscope at 40x magnification performed at the Otolith Lab at the Washington Department of Fish and Wildlife facilities in Olympia, Washington. The reference sample for wild-origin larvae was collected from the aforementioned emergence events using a combination of drift nets and fry emergence nets. Having captured the larvae the time they first emerged from their gravel nests, this guaranteed a control sample of larvae of 100% wild in rearing origin. Sagittal otoliths from euthanized Naches Fish Hatchery fry were removed on April 4th, 2020 in the aquatic laboratory at Central Washington University using the same methods as Gold Creek larvae. Larvae at the Naches Fish Hatchery began externally feeding on January

30th, 2020 and were kept in water temperatures fluctuating between 8.9 °C to 10 °C six to eight weeks before otolith extraction.

Larvae otoliths were removed using fine needle forceps and a thin wire probe. Otolith removal was performed with larvae dorsal side up. Skin was first removed from the skull, then the skull and brain were removed and discarded. Ethanol was added to the larvae in order to increase contrast between brain matter and otoliths. Extracted otoliths were washed in distilled water to remove any soft tissue and stored dry in glass vials prior to mounting. The left otolith for each larva were mounted onto a glass slide with a drop of thermoplastic resin (CrystalBond 509®). If the left otolith showed any damage or vaterite conformation of calcium carbonate the right otolith was then used instead. Mounted larvae otoliths were polished only on the sulcus side due to the otoliths at that age being semi-transparent. Polishing consisted of 5 µm aluminum oxide lapping paper combined with a distilled water and aluminum solution at approximately 1 µm grit. Otoliths were polished until the central primordia, hatch check, and exogenous feeding check were clearly visible.

Sagittal otoliths of kokanee salmon adults were removed using forceps and scalpel and prepared for microstructure examination in order to determine natal origins. Adult otolith preparation methods were the same as larvae otolith methods. Once otoliths were mounted, they were sanded wet on both sides, starting with 30 µm aluminum oxide lapping paper, then progressing to 15 and 5 µm grit. Adult otoliths were polished until the central primordia, hatch check, and exogenous feeding check were clearly visible.

Otolith Measurements

Once an otolith was sufficiently polished, microstructure features were identified and measured to determine differences in otolith structure between wild and hatchery-origin kokanee salmon. Locations of hatch and exogenous feeding checks for larvae and adults were determined by a consensus between myself and Dr. Wade Smith from the WDFW Otolith Lab (Figure 8; Figure 9). Images of otoliths were taken using Leica Microsystems® microscope imaging software under 10x, 20x, and 40x magnification. Images were taken periodically while otoliths were being polished to prevent the loss of distinguishing features. Images were analyzed using the assistance of free image analysis software ImageJ version 1.52a (Rasband, National Institutes of Health, USA) with the Measure and Label package.

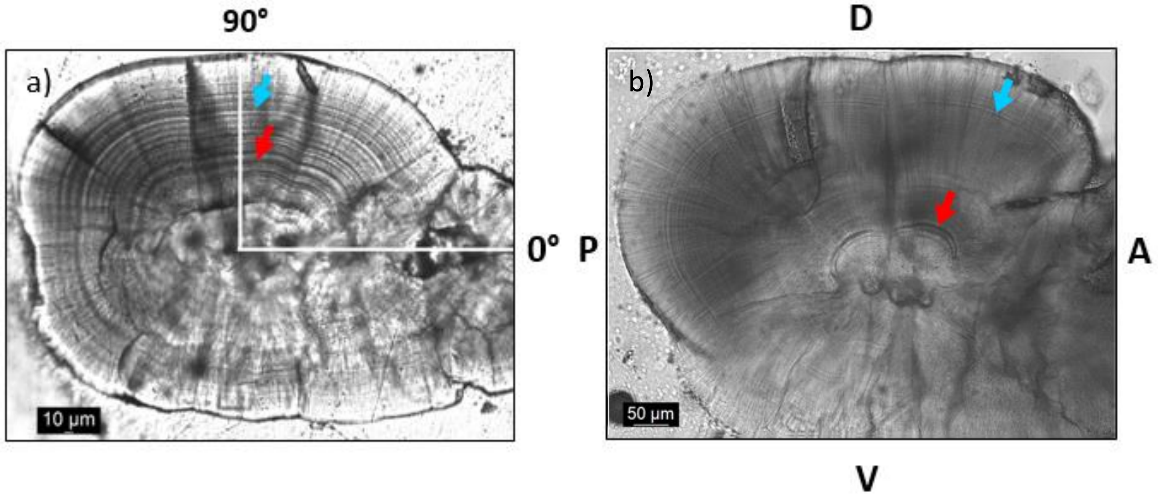

Figure 8. Image of a) wild-origin larva captured on April 28th, 2019 and b) Naches Fish Hatchery larva otoliths. Measurements of exogenous feeding check (blue arrow) and hatch check (red arrow) distances were taken using a ninety degree transect line located at the central primordia (D, dorsal; V, ventral; P, posterior; A, anterior).

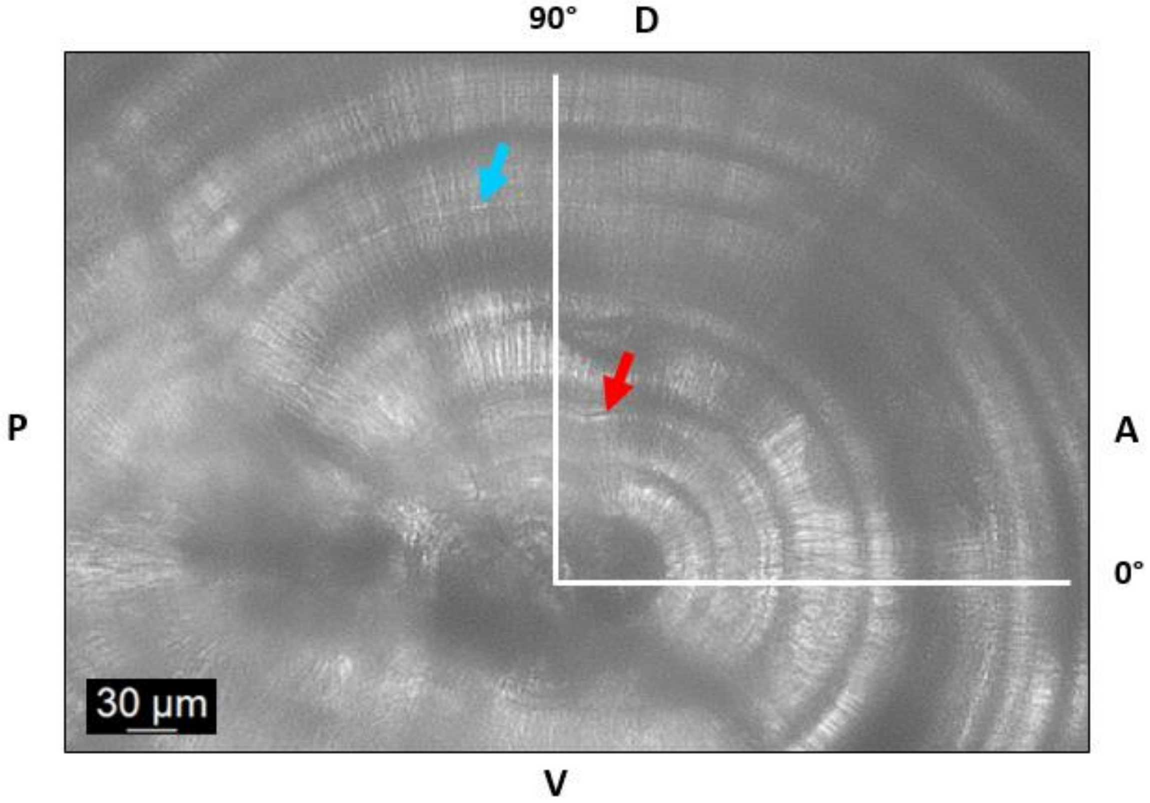

Figure 9. Image of a spawning adult kokanee otolith from the Gold Creek region collected fall 2013. Measurements of exogenous feeding check (blue arrow) and hatch check (red arrow) distances were taken using a ninety degree transect line located at the central primordia (D, dorsal; V, ventral; P, posterior; A, anterior)

In order to minimize variations in measurements, measurement standards were developed for adult and larvae otoliths. Distances to hatch and exogenous feeding checks for adult and larvae otoliths were measured following a transect 90° to the longest growth axis, crossing through the center of the primordia. Following suggestions from the studies of Zhang et al. (1995) and Barnett-Johnson et al. (2007), distances from the primordia to the hatch check, the primordia to the exogenous feeding check, and from the hatch check to the exogenous feeding check were measured. Measurements were always taken on the dorsal side of the otolith, chosen for uniform clarity. With these methods, I inferred that hatchery-origin adult otoliths would have a greater distance from the hatch check to the exogenous feeding check due to the fact that daily increments on otoliths were wider in hatchery salmonids compared to wild-origin salmonids (Barnett-Johnson et al. 2007).

Measurements from a subsample of five larvae otoliths from wild-origin and hatchery-origin larvae reference samples were taken by an additional reader, with a consensus being reached if measurements were within five µm of each other. In the case of disagreement, both readers would measure the otolith until agreement. For adult otoliths, I measured distances and then a subsample was again measured by an additional reader without prior knowledge of the rearing origin of adults. A consensus was reached if measurements were within five µm of each other. In the case of disagreement, both readers would measure the otolith again until measurements were within the acceptable range.

Statistical Analysis

Size

A Kruskal-Wallis rank sum test was used to compare larvae total lengths due to the continuous variable not meeting the assumptions of normality and homogeneity of variances for one-way analysis of variance (ANOVA) tests. An ANOVA was used to compare larvae widths between sample locations in the Gold Creek ecosystem from April 25th to May 29th, 2019, with the continuous variable meeting the assumptions of normality and homogeneity of variances. Sample locations were above the beaver dam (Above) and below the beaver dam (Below) in the Gold Creek Pond outlet channel, emergence net 2 (Enet2) directly upstream of the beaver dam, emergence net 3 (Enet3) upstream of the beaver dam near Gold Creed Pond, and in Gold Creek by the I-90 bridges.

Beaver Dam Effects on Larvae

Differences in larvae densities between sample locations (above and below the beaver dam in the Gold Creek Pond outlet channel) were analyzed using a nonparametric Mann-Whitney U Test due to the data not meeting assumptions of normality for a Welch two-sample t-test. Differences in larvae density by collection date were plotted alongside changes in mean groundwater temperature to show the average water temperature needed for kokanee larvae to start emerging.

Larvae Otolith Microstructure Analysis

A Welch two-sample t-test was used to compare distances (µm) from the central primordia to the hatch check, from the central primordia to the exogenous feeding check, and from the hatch check to the exogenous feeding check for hatchery-origin larvae versus wild-origin larvae. The continuous variables all met the assumptions of normality for a Welch two-sample t-test.

Adult Otolith Microstructure Analysis

A linear discriminant analysis (LDA) was performed on the wild and hatchery known origin larvae dataset in order to determine whether the otolith measurements could be used to correctly classify individual kokanee salmon by rearing origin. All three continuous response variables met the assumptions of normality for a LDA. The minimum and maximum values from the LDA for wild and hatchery-origin larvae were calculated. Otolith measurements of unknown-origin adults were then quantified using the LDA model. LDA values of unknown-origin adults were compared to the minimum and maximum LDA values for wild-origin and hatchery-origin larvae in order to assign most likely rearing origin.

A Welch two-sample t-test was used to compare distances (µm) between the central primordia and the hatch check, the central primordia and the exogenous feeding check, and the hatch check and the exogenous feeding check for predicted hatchery-origin adults versus predicted wild-origin adults. The continuous variables primordia to hatch check distance and primordia to exogenous feeding check distance met the assumptions of normality for a Welch two-sample t-test. The continuous variable hatch check to exogenous feeding check distance met the assumption of normality after a log transformation. All statistical analyses were performed in R (Version 1.3.1093).

CHAPTER IV

RESULTS

Larvae Emergence Timing and Size

Kokanee salmon were observed spawning from mid-September to mid-November 2018. Redds were observed monthly from February 2019 to March 2019 to monitor egg development. Viable eggs (eyespot present) were found in the gravel beds until the third week of April 2019. Alevin were observed from periodic sampling until April 7th, 2019. Young-of-year were first observed in the study site on April 25th, 2019. Prior to this date I returned to Gold Creek every other day in the two weeks leading up to the first emergence date to check emergence development. Larvae emergence occurred until May 29th, 2019, with a total emergence time period lasting 34 days. The observed larvae emerged approximately six months after redds were built by spawning adults the previous fall (Figure 10).

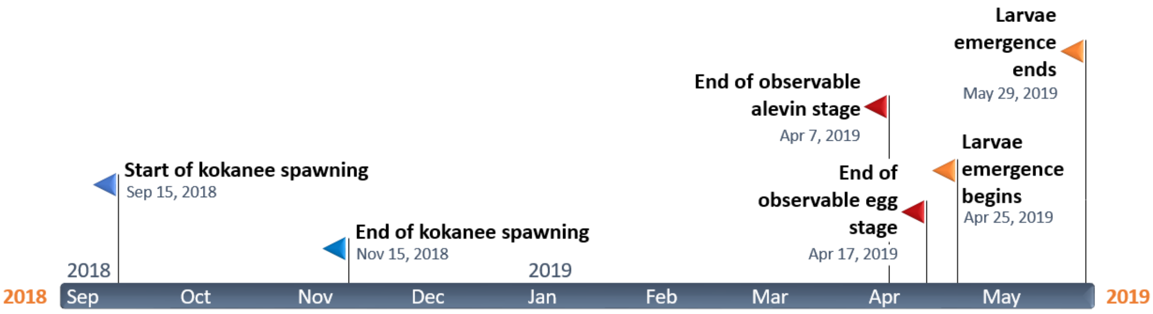

Figure 10. Timeline of adult kokanee salmon spawning period and subsequent young-of-year life stages in the Gold Creek ecosystem.

The total number of larvae observed emerging from Gold Creek and the Gold Creek Pond outlet channel was 323 captured using drift nets and six using fry emergence nets. In the Gold Creek Pond outlet channel, 280 larvae were observed emerging above the beaver dam compared to 22 larvae below the beaver dam. In Gold Creek itself, four larvae were captured using drift nets in the creek adjacent to the I-90 bridges and only one larva was captured in Gold Creek near the confluence of Gold Creek and the Gold Creek Pond outlet channel (Figure 11).

For each larva, total length was measured from April 25th, 2019 to May 29th, 2019. Biweekly total length measurement averages were calculated during this time period (Table 2). Average lengths increased by approximately 2 mm in larvae emerging at the end of the emergence event compared to larvae emerging at the beginning. Average width of larvae did not differ over their emergence period. All larvae captured were assessed at a development level of five, having entirely absorbed their maternal yolk sacs.

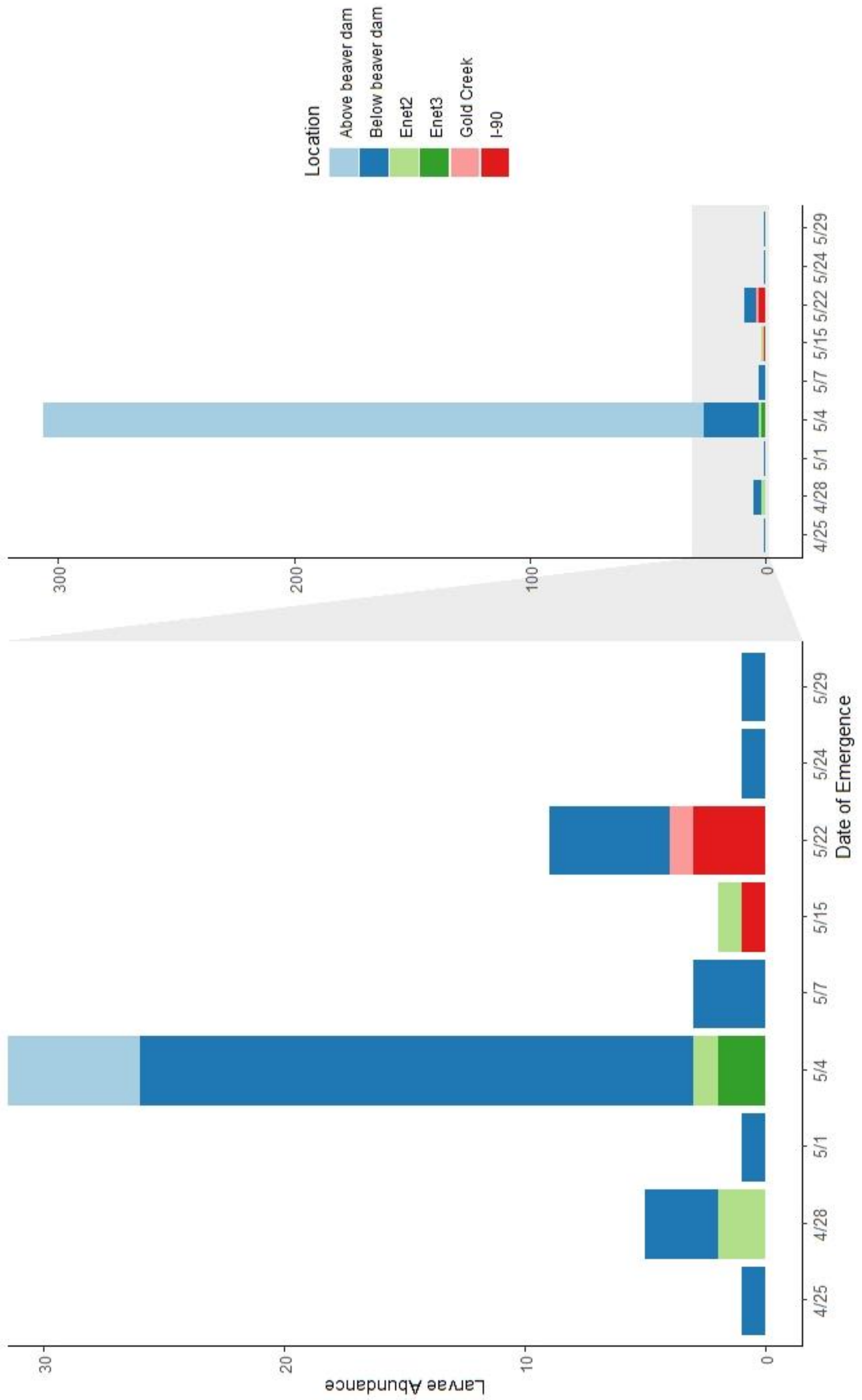

Figure 11. Abundance of larvae on date of emergence described by location within the Gold Creek ecosystem in the spring of 2019. Larvae abundance peaked on May 4th, 2019.

Table 2. Average total lengths and widths of larvae taken biweekly from April 25th to May 29th, 2019.

Emergence Date	**# of Individuals**	**Mean Total Length (mm)**	**Mean Width (mm)**
April 25th to May 1st, 2019	4	24	2.9
May 1st to May 14th, 2019	47	26.3	3.0
May 15th to May 29th, 2019	12	26.3	3.1

The length of larvae did not differ significantly between sample locations, with Gold Creek and emergence net 1 sample locations not included in the analysis due to low sample size (Figure 12, $p > 0.05$; Kruskal-Wallis rank sum test). The width of larvae also did not differ significantly between sample locations, with Gold Creek and emergence net 1 sample locations being excluded as well (Figure 13, $p > 0.05$; one-way ANOVA test).

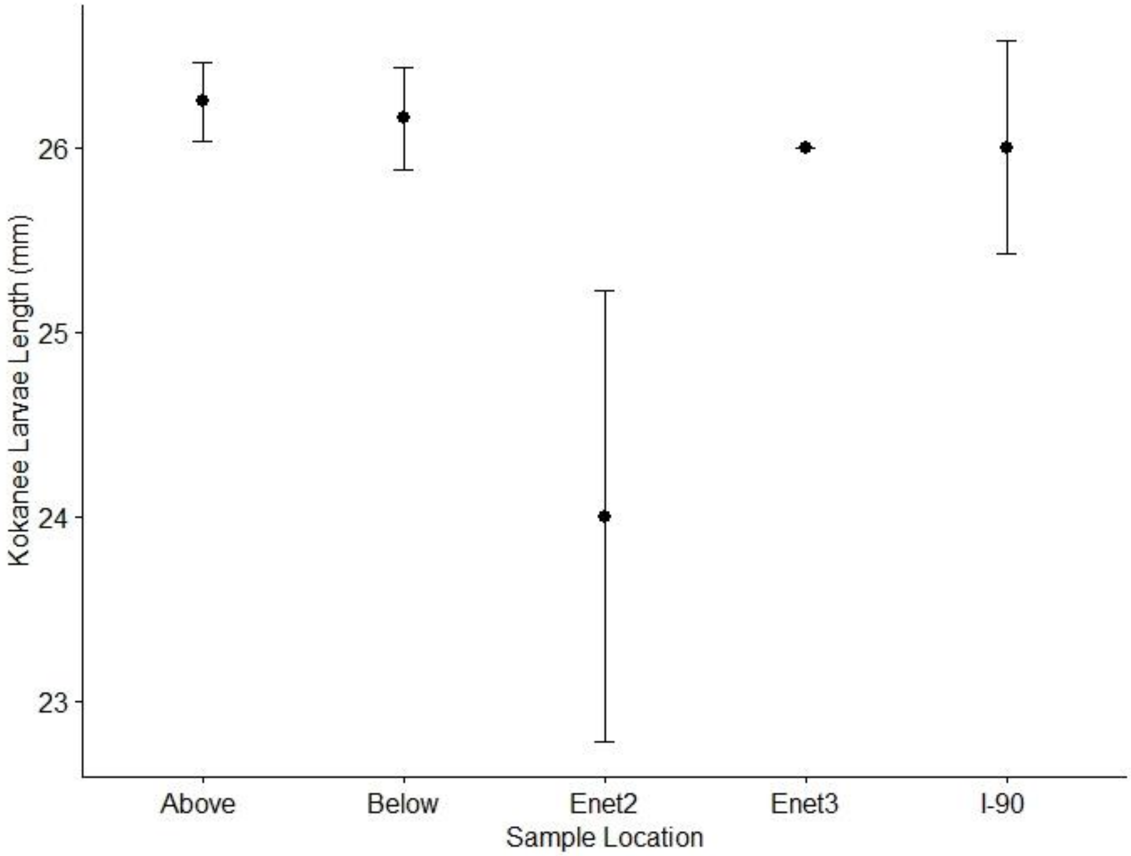

Figure 12. Comparison of larvae total length measurements (mm) between sample locations (above and below the beaver dam in the Gold Creek Pond outlet channel, emergence net 2 and 3 in the Gold Creek Pond outlet channel, and Gold Creek downstream of the I-90 bridges) in the Gold Creek ecosystem between April 25th and May 31st, 2019. Error bars represent standard error of the mean.

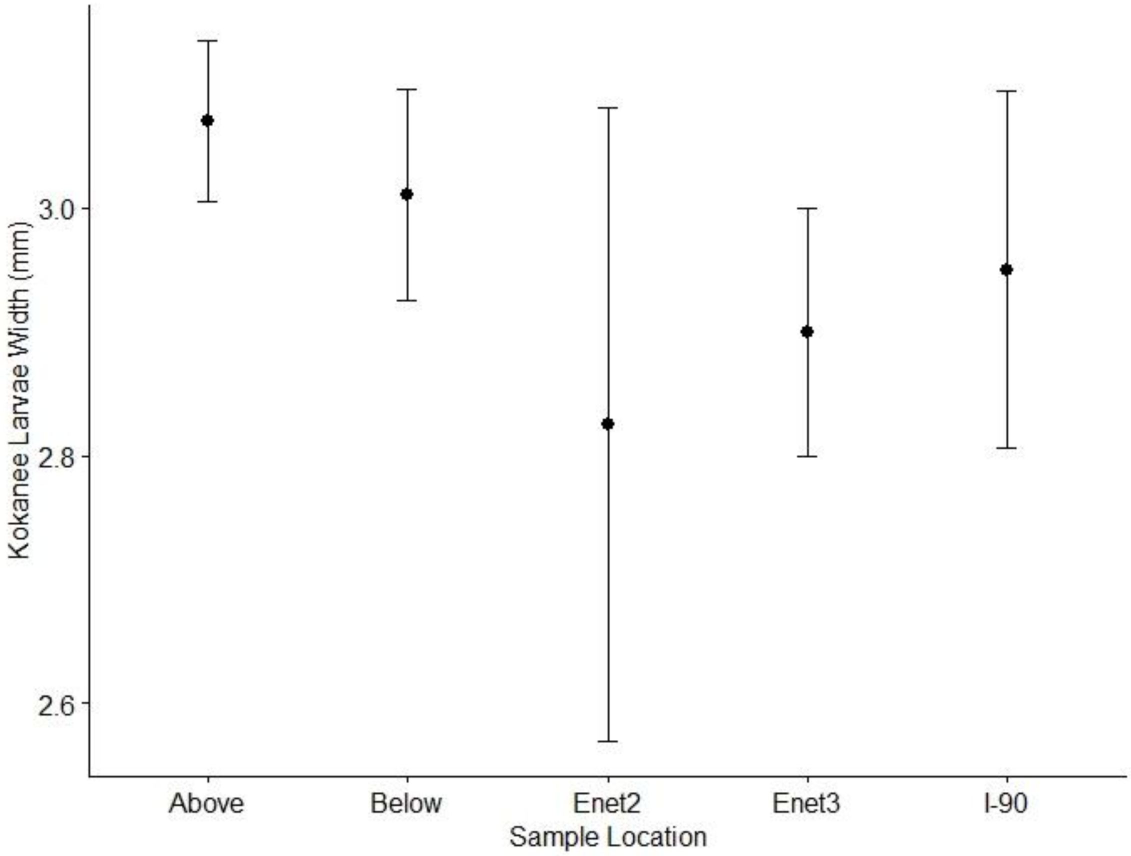

Figure 13. Comparison of larvae width measurements (mm) between sample locations (above and below the beaver dam in the Gold Creek Pond outlet channel, emergence net 2 and 3 in the Gold Creek Pond outlet channel, and Gold Creek downstream of the I-90 bridges) in the Gold Creek ecosystem between April 25th and May 31st, 2019. Error bars represent standard error of the mean.

Beaver Dam Effects on Larvae

Above and below the beaver dam in the Gold Creek Pond outlet channel had the only significant difference in larvae density. A significantly higher density of larvae emerged below the beaver dam than above the beaver dam between April 25th and May 29th, 2019 (Figure 14, $p = 0.008$, Mann-Whitney U Test). The average density of larvae above the beaver dam was zero larvae/100m^3 not counting outliers, 1.76 larvae/100m^3

counting outliers, and the average density of larvae below the beaver dam was 0.46 larvae/100m^3.

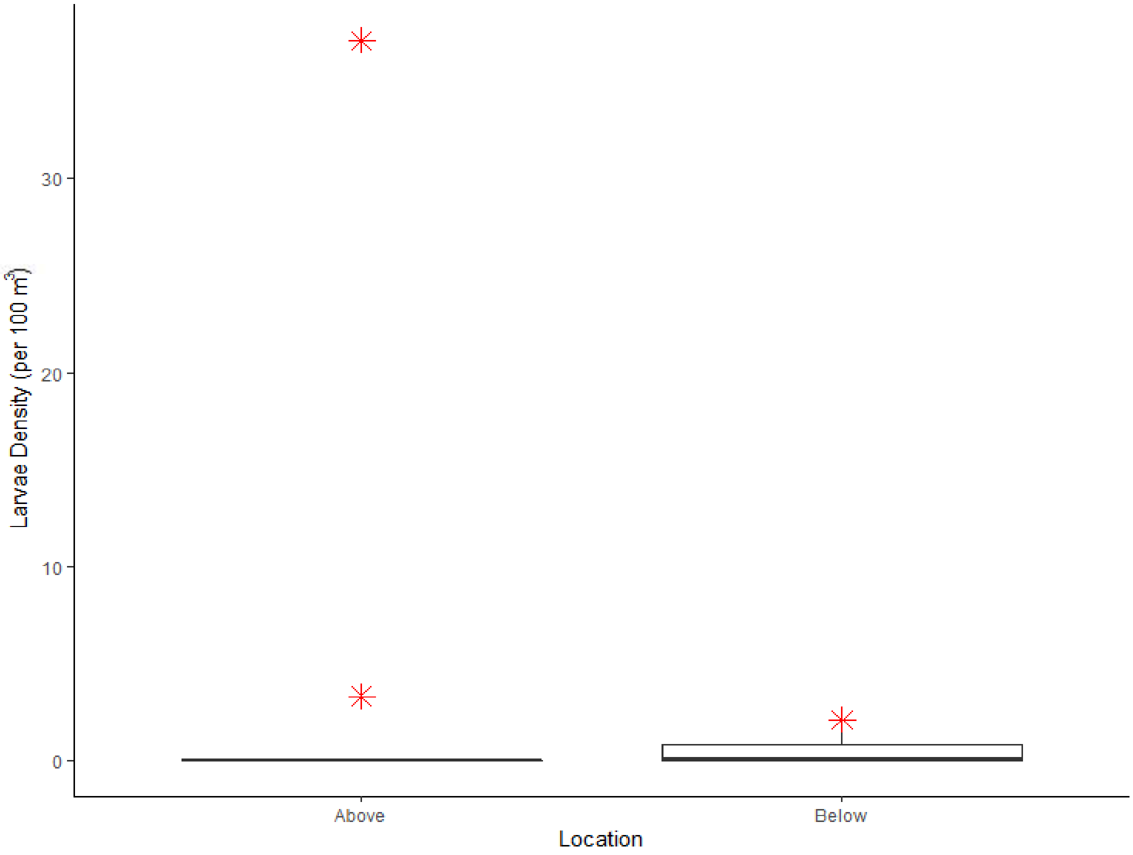

Figure 14. Comparison of the larvae density (number of larvae/100 m^3) above and below the beaver dam in the Gold Creek Pond outlet channel.

The average groundwater temperature of Gold Creek at the start of larvae emergence was 3.3 °C (Figure 15). Mean surface water temperature of Gold Creek at the beginning of larvae emergence, April 25th, 2019, was 3.4 °C and ambient air temperature had a minimum of -2.2 °C and a maximum of 16.9 °C (Snoqualmie Pass weather station).

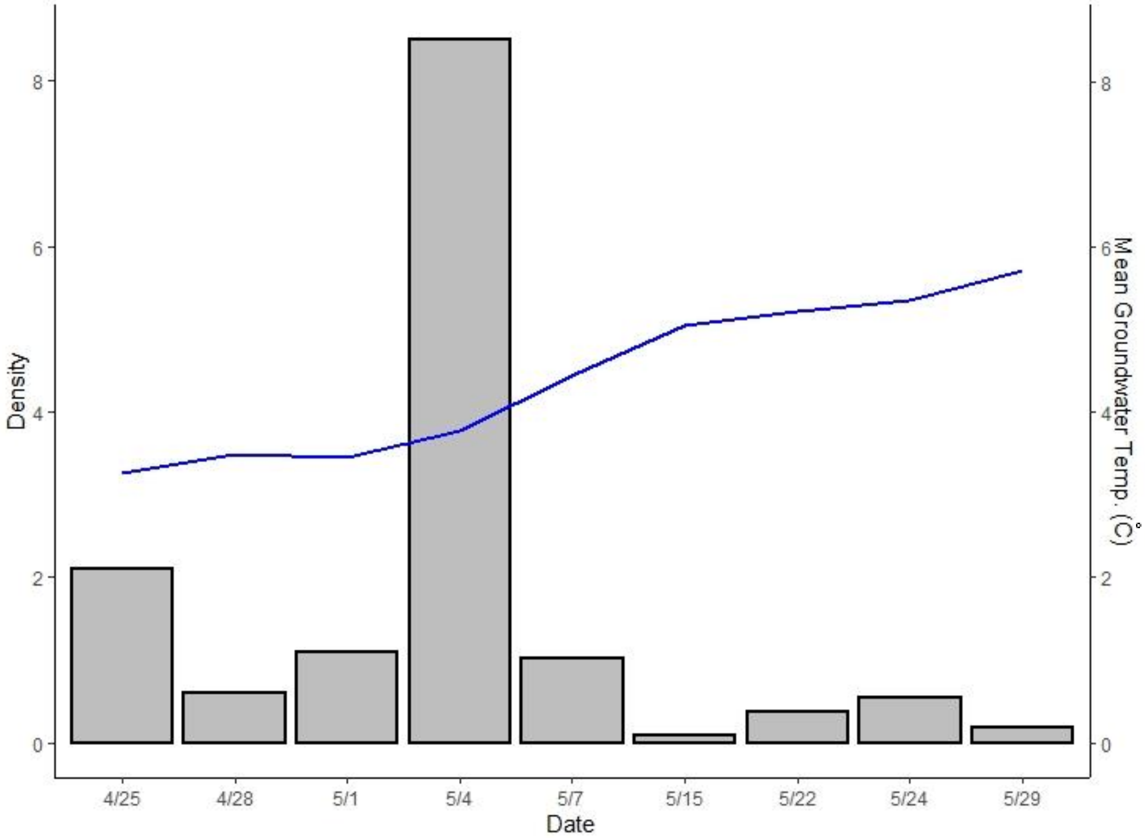

Figure 15. Mean density (number of larvae/100 m^3) of emerged larvae described by date of collection with daily groundwater temperature (°C) of Gold Creek in 2019.

Larvae Otolith Microstructure Analysis

To compare otolith microstructure differences between wild and hatchery-origin larvae, 18 Gold Creek wild-origin and 20 Naches Fish Hatchery origin larvae were used (example shown in Figure 16). Hatchery-origin larvae had significantly shorter distances between the central primordia and the hatch check compared to wild-origin larvae (Figure 17, two sample t = -5.336, df = 33.728, p < 0.001). Hatchery-origin larvae had significantly larger distances between the central primordia and the exogenous feeding check compared to their wild-origin counterparts (Figure 18, two sample t = 10.408, df = 35.703, p < 0 .001). In addition, hatchery-origin larvae had significantly larger distances between the hatch check and the exogenous feeding check compared to wild-origin larvae (Figure 19, two sample t = 15.698, df = 35.724, $p < 0.001$). Independent validation of larvae measurements by an additional reader was in consensus except for one hatchery larva hatch check to exogenous feeding check measurement, where an additional measurement by both readers was needed to reach a consensus.

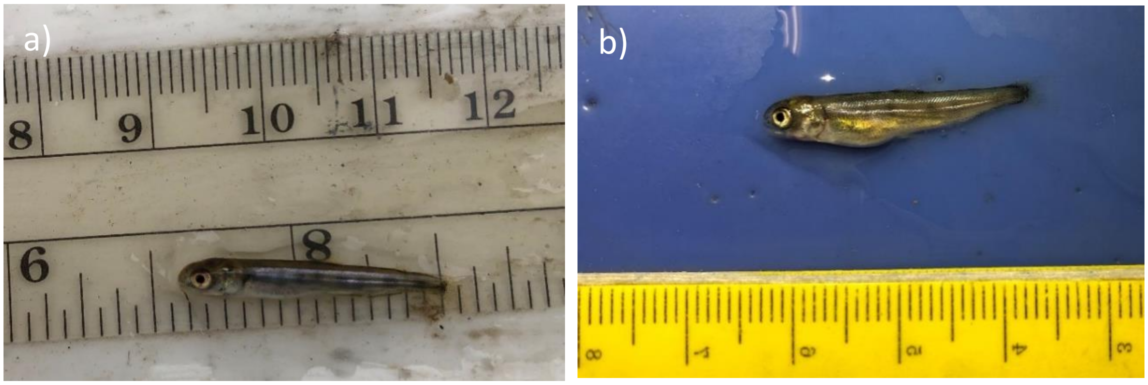

Figure 16. Photographs of young-of-year kokanee salmon taken a) newly emerged from the Gold Creek Pond outlet channel on May 24th, 2019 and b) from the Naches Fish Hatchery on February 25, 2020.

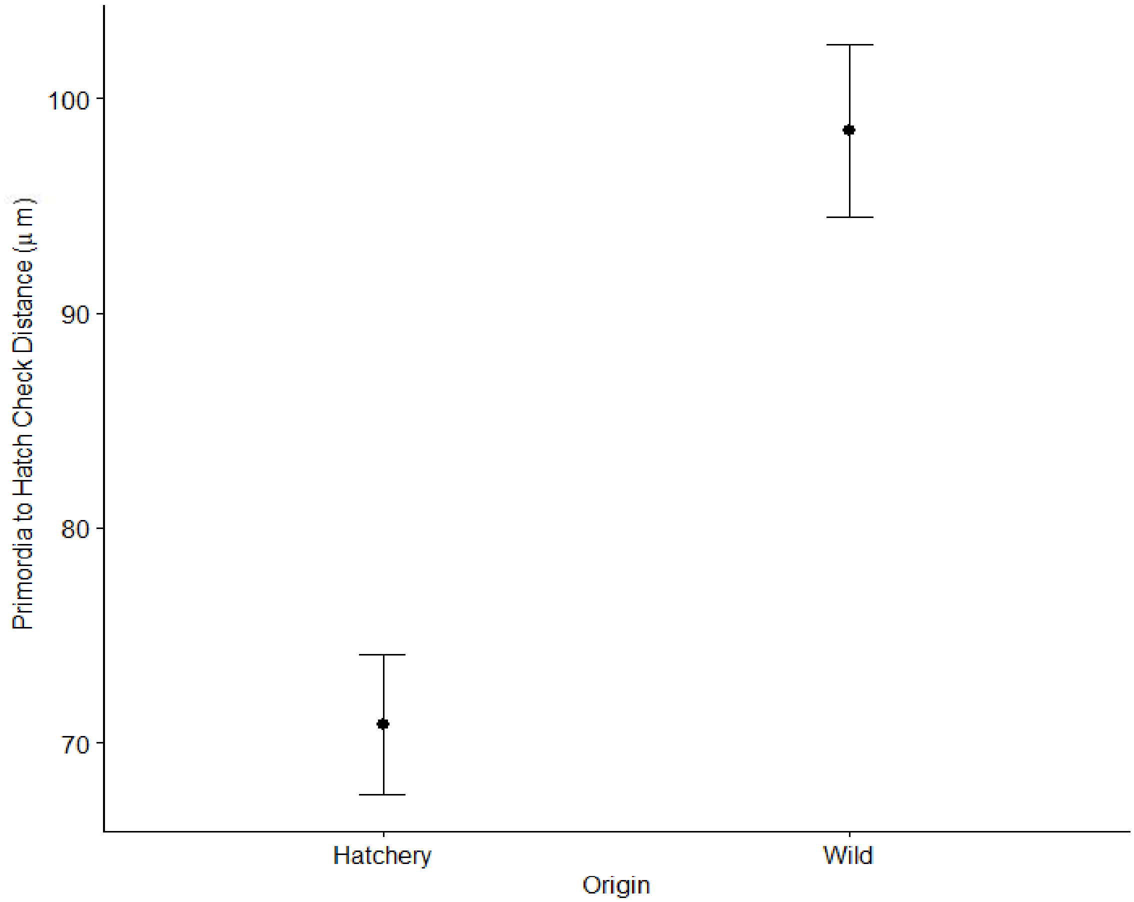

Figure 17. Comparison of larvae otolith distances (µm) from the central primordia to the hatch check between hatchery-origin and Gold Creek wild-origin larvae. Error bars represent standard error from the mean.

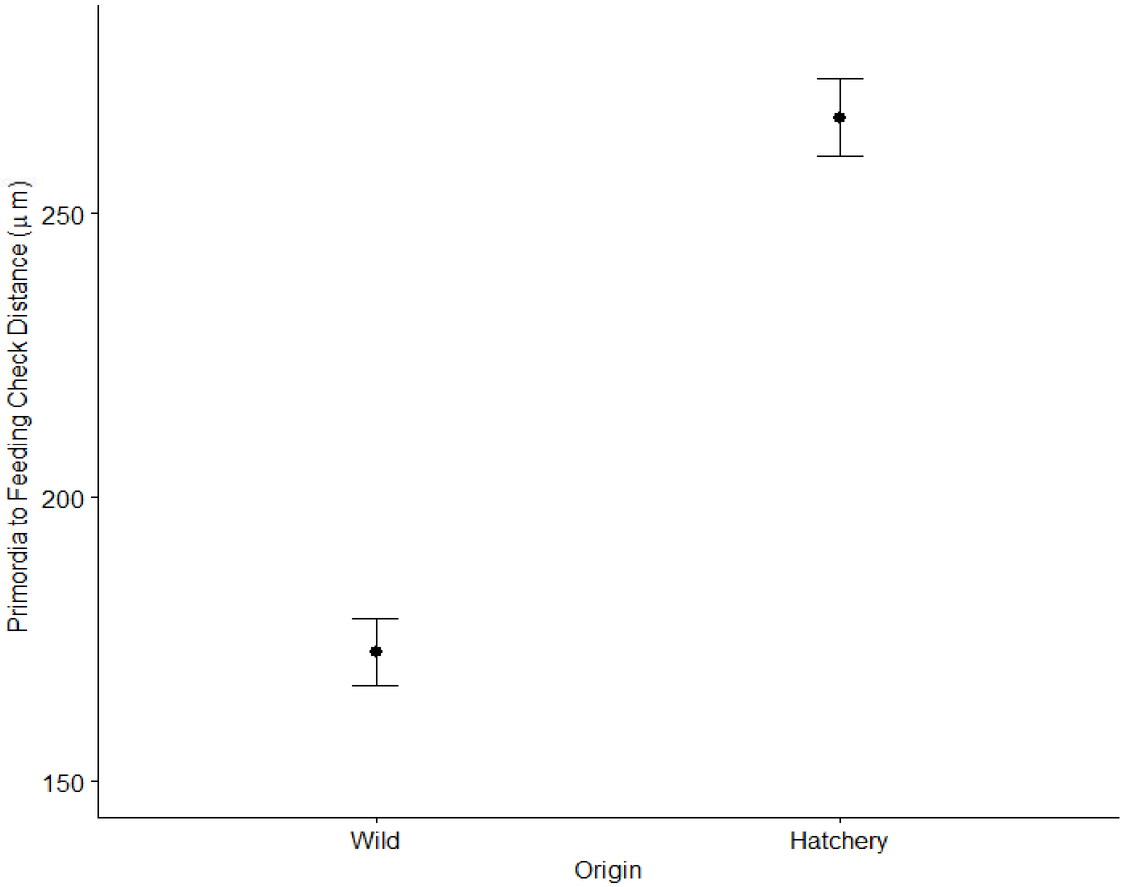

Figure 18. Comparison of larvae otolith distances (µm) from the central primordia to the exogenous feeding check between hatchery-origin and Gold Creek wild-origin larvae. Error bars represent standard error from the mean.

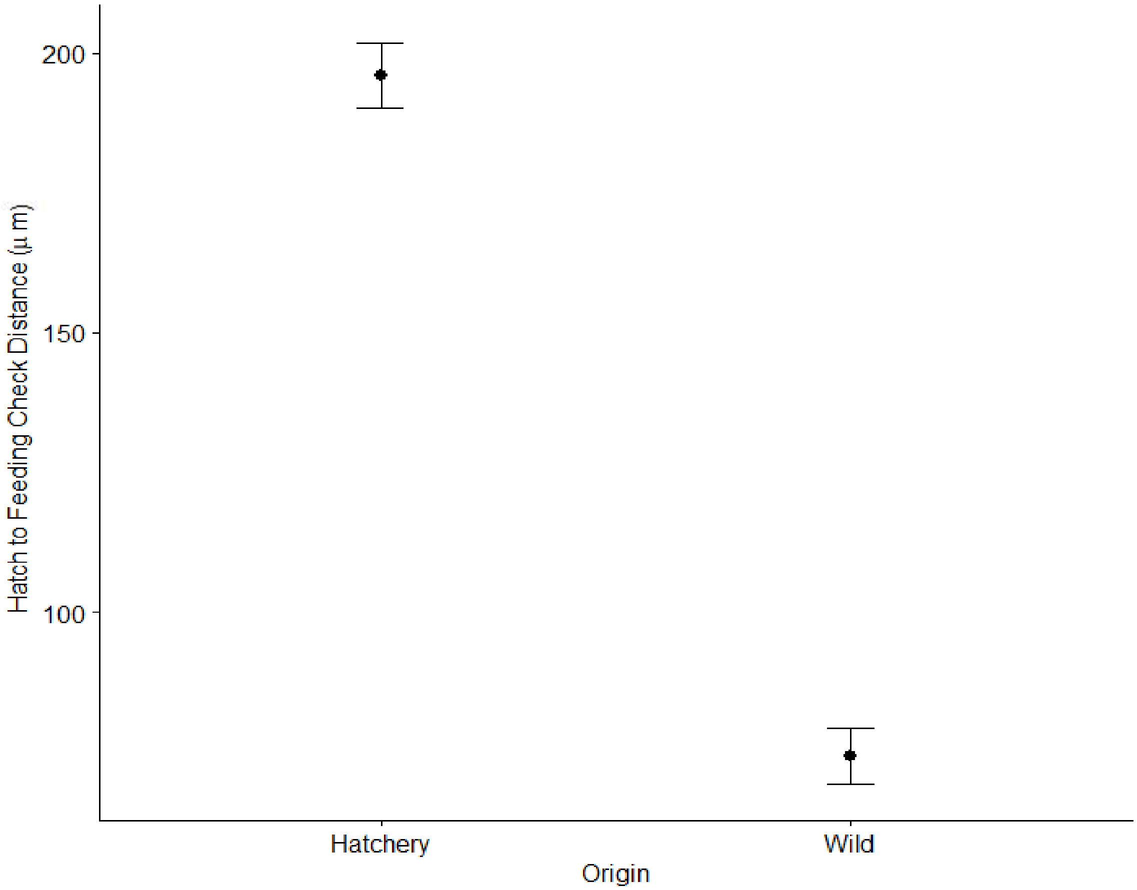

Figure 19. Comparison of larvae otolith distances (µm) from the hatch check to the exogenous feeding check between hatchery-origin and Gold Creek wild-origin larvae. Error bars represent standard error from the mean.

The LDA values resulting from the otolith measurements for both hatchery and wild larvae were plotted using a histogram in order to determine the minimum and maximum LDA values and to check for a satisfactory separation of values between the two groups. A distinct separation in LDA values was observed for wild and hatchery larvae of known rearing origin (Figure 20). The mean LDA value for wild-origin larvae was -1.16 with a minimum value of -3.83 and a maximum value of 0.42. The mean LDA value for hatchery-origin larvae was -6.42 with a minimum value of -8.95 and a maximum value of -5.07.

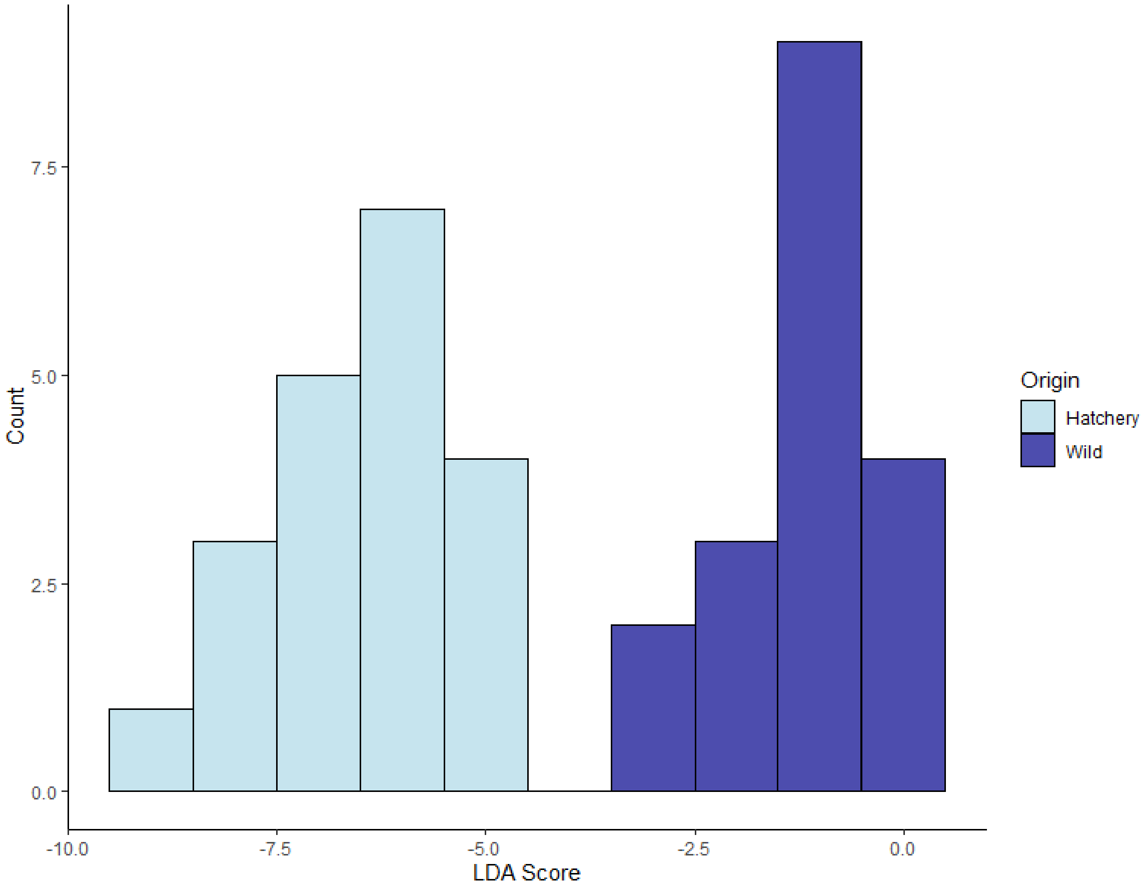

Figure 20. Histogram of LDA values for the observations of hatchery-origin and wild-origin larvae groups.

The LDA model using known-rearing-origin larvae produced the following linear equation

$$LDA = -0.01129963(x) + 0.0290597(y) -0.02786657(z)$$

where x is the distance (µm) from the central primordia to the hatch check, y is the distance (µm) from the central primordia to the exogenous feeding check, and z is the distance (µm) from the hatch check to the exogenous feeding check.

Adult Otolith Microstructure Analysis

Using the LDA model from the larval otolith measurements, parameters for unknown origin adults were used to produce LDA values that were then compared to the LDA values of known hatchery-origin and wild-origin larvae. Any adult LDA value greater than -3.406 was classified as wild origin and any LDA value less than -3.406 was classified as hatchery origin. A separation in LDA values was observed for predicted wild-origin and predicted hatchery-origin larvae (Figure 21). The mean LDA value for predicted wild-origin adults was -1.36 with a minimum value of -3.36 and a maximum value of 1.24. The mean LDA value for predicted hatchery-origin adults was -5.09 with a minimum value of -9.69 and a maximum value of -3.56. Results from the LDA model using known-origin kokanee larvae suggest that 50 out of 60 adult kokanee salmon, or 83%, most likely have wild origins.

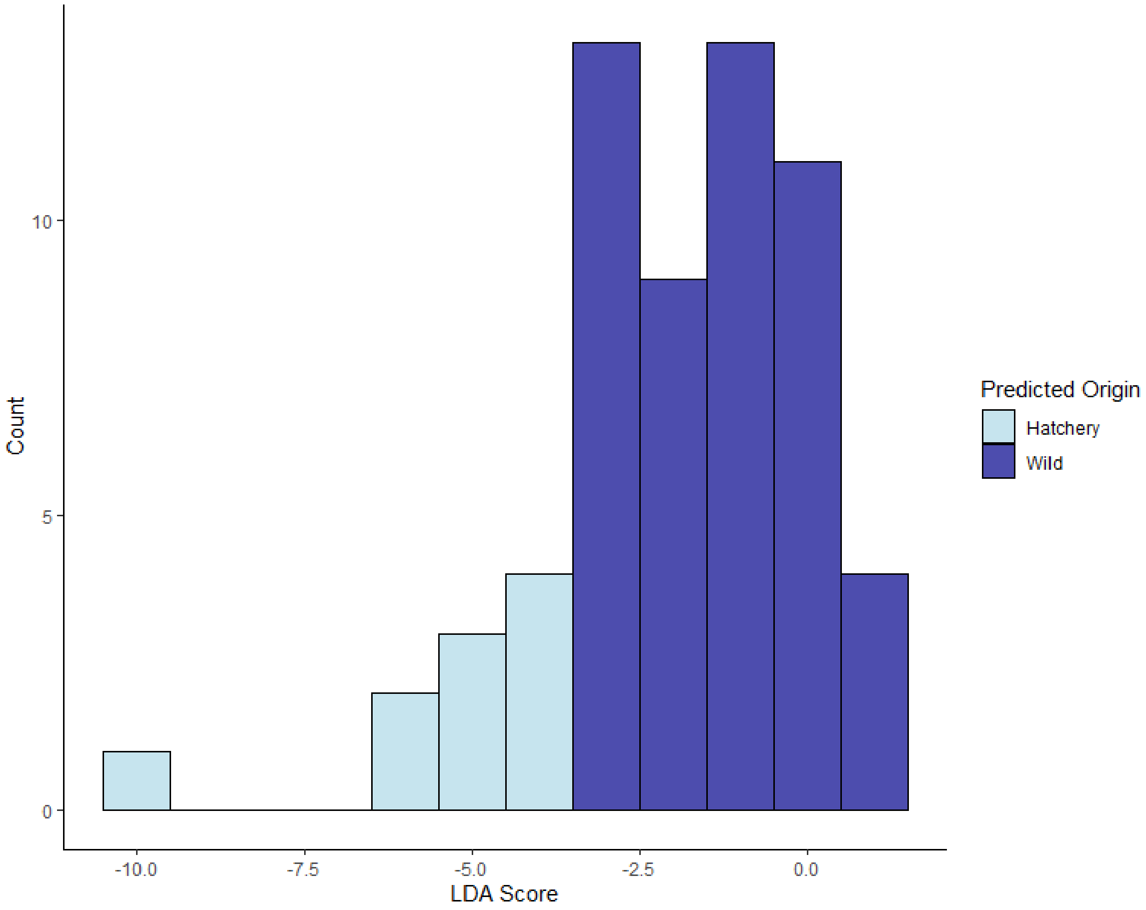

Figure 21. Histogram of LDA values for the observations of predicted wild-origin and predicted hatchery-origin adult groups.

Based on LDA classification, otolith microstructure differences were compared between wild and hatchery-origin adults. Hatchery-origin adults did not have significantly different distances between the central primordia and the hatch check compared to wild-origin adults (Figure 22, two sample t = -1.899, df = 15.978, p = 0.0758). Hatchery-origin adults had significantly larger distances between the central primordia and the exogenous feeding check compared to their wild-origin counterparts (Figure 23, two sample t = 2.739, df = 10.793, p = 0.020). In addition, hatchery-origin adults had significantly larger distances between the hatch check and the exogenous feeding check compared to wild-origin adults (Figure 24, two sample t = 7.173, df = 17.875, $p < 0.001$).

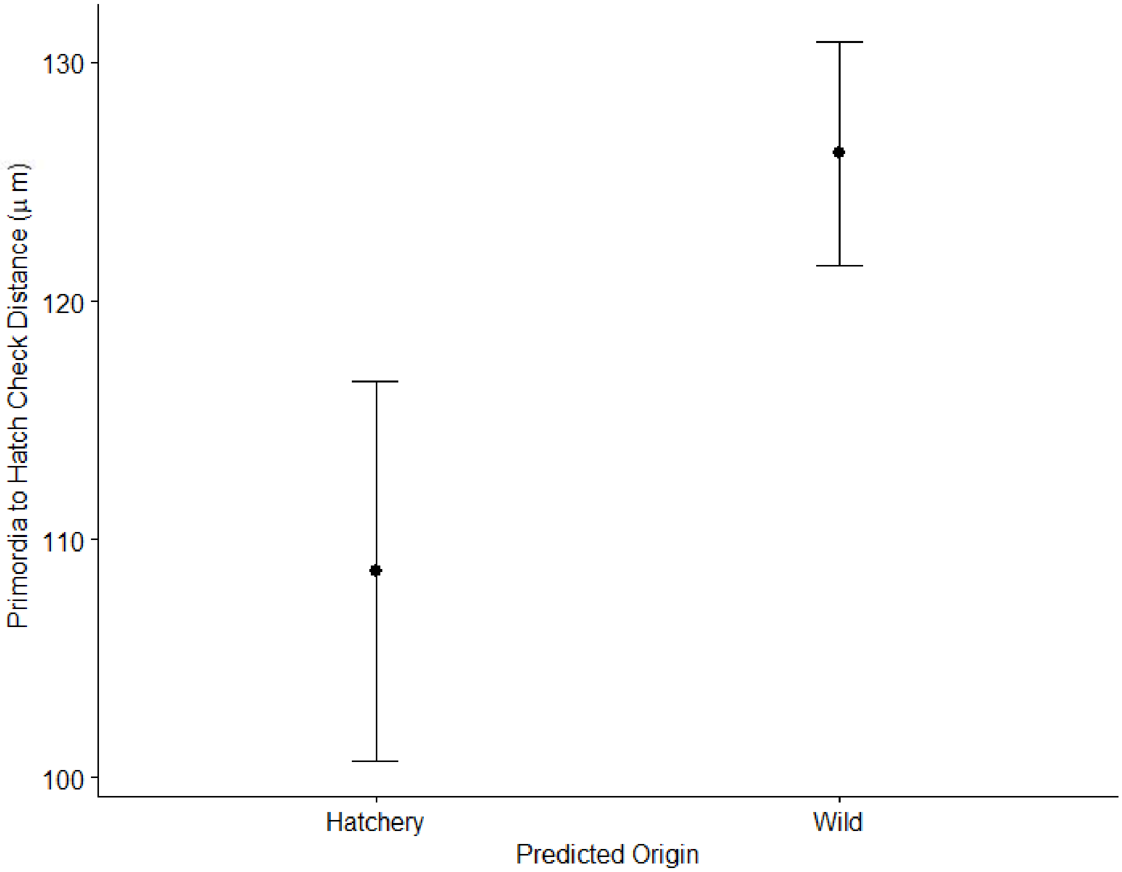

Figure 22. Comparison of otolith distances (µm) from the central primordia to the hatch check between predicted hatchery-origin and predicted Gold Creek wild-origin adults. Error bars represent standard error from the mean.

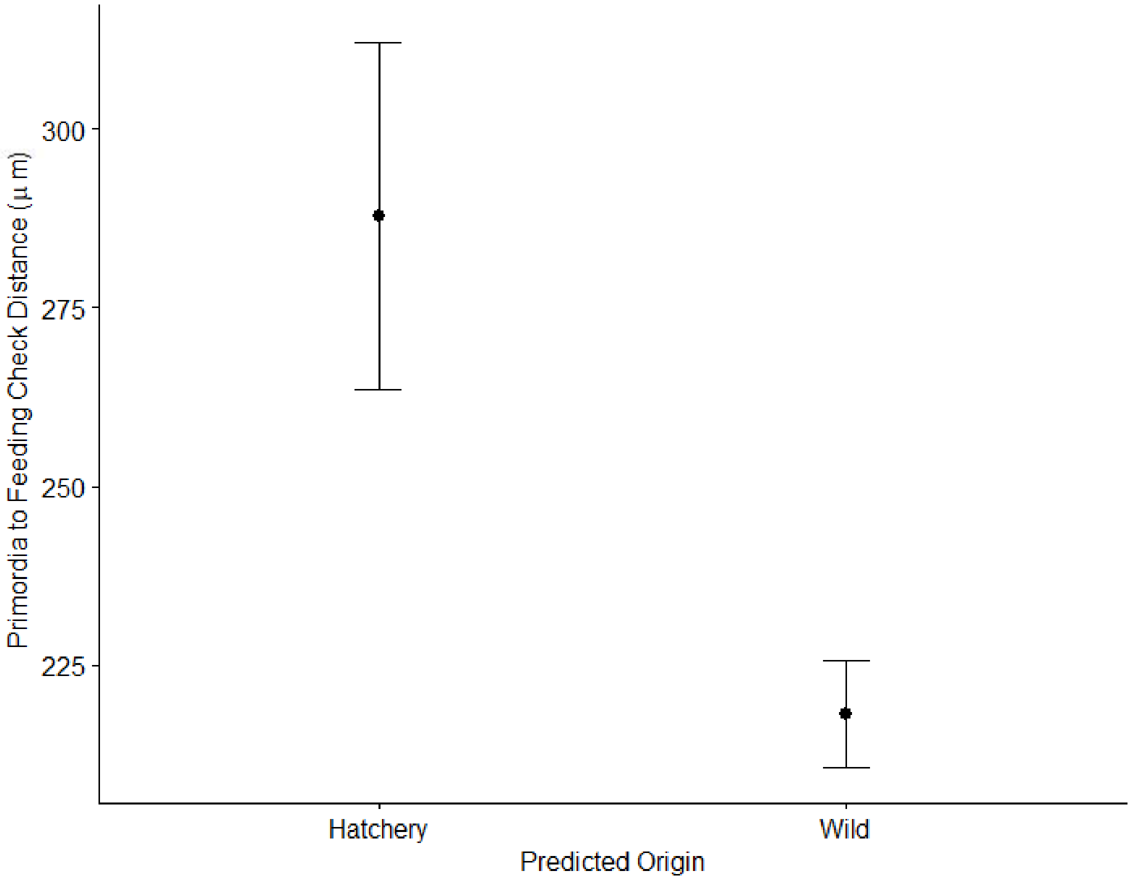

Figure 23. Comparison of otolith distances (µm) from the central primordia to the exogenous feeding check between predicted hatchery-origin and predicted Gold Creek wild-origin adults. Error bars represent standard error from the mean.

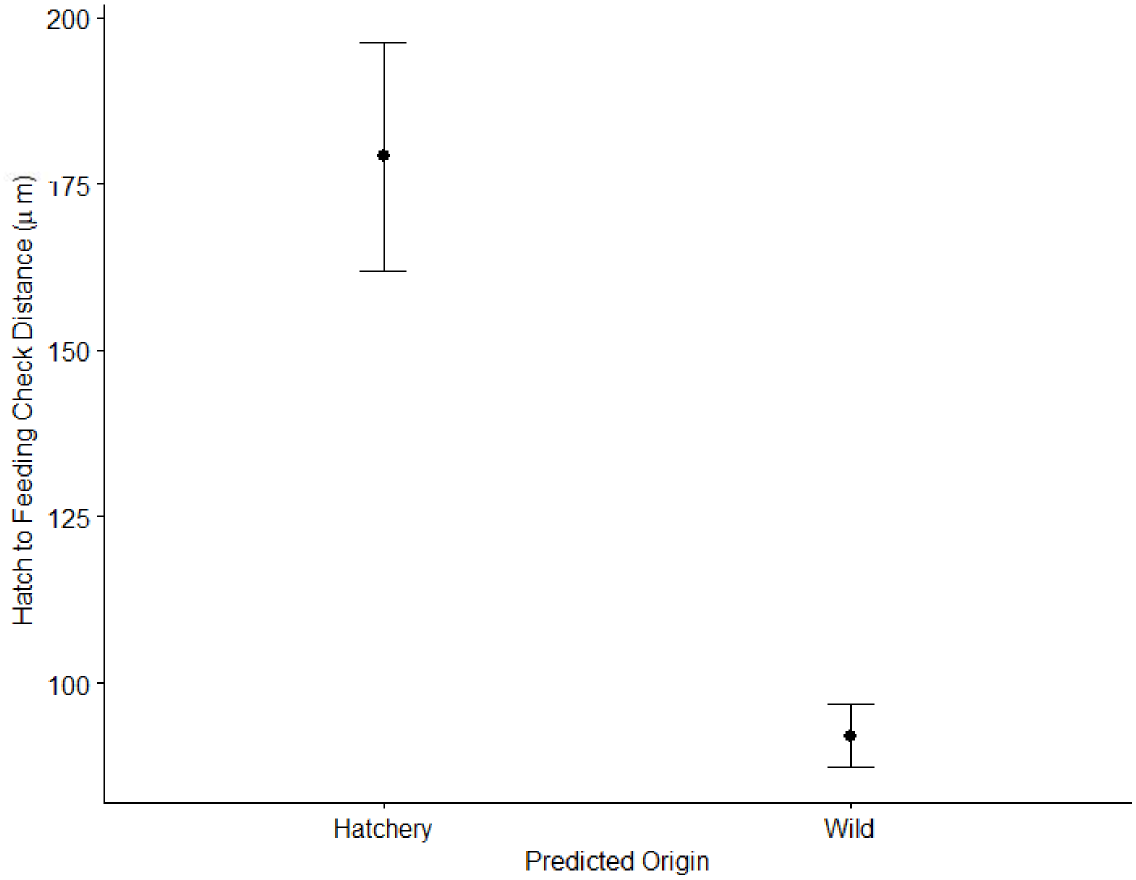

Figure 24. Comparison of otolith distances (µm) from the hatch check to the exogenous feeding check between predicted hatchery-origin and predicted Gold Creek wild-origin adults. Error bars represent standard error from the mean.

CHAPTER V

DISCUSSION

To my knowledge, this is the first study of its kind analyzing the early life history of kokanee salmon in an alpine lake environment in Washington State. Kokanee salmon, being a popular recreational fishing species and frequently raised in hatcheries, has been the center of numerous studies. These studies, however, tend to focus on thermal marking strategies for hatchery kokanee salmon (Paragamian et al. 1992), the impact of anadromous sockeye salmon mixing with kokanee salmon (Veale and Russello 2016), the introduction of kokanee salmon to previously unoccupied ecosystems (Hansen et al. 2016; Lyons et al. 2019), and the evolution and population structures of the kokanee “resident” ecotype (Lemay et al. 2013; Lemay and Russello 2015; Beacham and Withler 2017; Veale and Russello 2017). No studies, however, focused solely on a descriptive early life history of kokanee salmon in their natal environment.

Prior to the start of my research project, knowledge of the kokanee salmon population that resides in Keechelus Lake ended with the fall spawning of adults in Gold Creek. Approximately 300,000 hatchery sourced kokanee salmon (Thompson 2019) are being stocked annually into an ecosystem where no one knew if kokanee salmon were successfully spawning and if their potential offspring could survive the winter and emerge in the spring. After extensive monitoring using a combination of drift and fry emergence nets, I successfully documented the development of larvae through the winter and spring of 2019, showing that adults that spawn in the Gold Creek ecosystem have a self-sustaining life cycle that results in viable young-of-year. In addition, the early life history and development of the larvae were evaluated in regards to the presence of a well-established beaver dam and I-90 bridges. High densities of adult spawners and subsequent larvae offspring in the Gold Creek Pond outlet channel compared to Gold Creek itself suggests significant barriers to kokanee salmon spawning dispersion and sheds light onto ongoing ecosystem challenges in Gold Creek.

Emergence Timing

The emergence timing of larvae occurred later than what was predicted. Larvae were first observed emerging in the Gold Creek Pond outlet channel below the beaver dam starting on April 25th, 2019. Emergence lasted until May 29th, 2019, spanning 34 days. On average the eggs deposited in redds in the Gold Creek ecosystem emerged in 200 days, with the average spawning date being October 15th, 2018 and the average emergence date being May 12th, 2019. The emergence of Gold Creek larvae does not correspond to reported young-of-year kokanee salmon average length of emergence. Brown, C.J.D. (1971) reported kokanee salmon eggs, with wild-rearing origins in Montana, hatch after an average of 110 days and emerge after 140 days at 6.1 °C, usually between March and early April. Meanwhile research by Stober and Tyler (1981) studying kokanee salmon spawning in Banks Lake, Washington, concluded that adults that spawned between the months of October and November had young-of-year emerge from late March to early June. This reported incubation time before emergence of approximately 150 days matches closer to the observed incubation time of Gold Creek larvae.

These discrepancies in emergence lengths relate to numerous factors, such as changes in thermal regimes, whether spawning was on lake shores or in lake tributaries, and depth of redds (Stober and Tyler 1981). In the case of Gold Creek and the Snoqualmie Pass region, the winter of 2018/2019 had an unusually late snow season, with the last measurable snow fall recorded on April 14th, 2019 with peak snowfall occurring mid-February. The later timing of larvae emergence could be a result of a heavy snowpack during the early spring months when emergence, according to literature, normally occurs. However, there is a distinct lack of scientific studies regarding exact timing of kokanee salmon egg incubation lengths and emergence timing in a non-hatchery setting. Additional studies are recommended to further analyze potential emergence times of kokanee salmon in their natal environment, especially regarding adults that spawn in the Gold Creek ecosystem.

For future management implications, the utilization of habitat parameters and emergence timing data from this study can be useful to predict future larvae emergence in the Gold Creek ecosystem. For the larvae that first started emerging on April 25th, 2019, water temperature benchmarks of 3 °C to 4 °C and air temperatures ranging from a minimum of 2 °C to a maximum of 17 °C can be used for future estimations of larvae emergence. In the case of future restoration efforts in the Gold Creek ecosystem, I recommend halting projects between mid-April to the first week of June in order to prevent harming newly emerged young-of-year until they safely exit the Gold Creek reach and enter Keechelus Lake.

Beaver Dam Effects

The presence of the beaver dam in the Gold Creek Pond outlet channel had no significant effect on either the length or width of the larvae that developed in its presence. There was a significantly higher number of larvae that emerged above the beaver dam, 280 larvae, compared to the 22 larvae that were observed emerging below the beaver dam. The high abundance of larvae upstream of the beaver dam, and numerous personal observations of adults finding passages underneath the dam to gain access to the reach above, indicates that the beaver dam itself was not a barrier to spawning kokanee salmon in the fall. In contrast to the larger abundance of larvae above the beaver dam, density of larvae was highest below the beaver dam. A mass exodus of larvae on May 4th, 2019, with 256 larvae captured in two separate drift nets, contributed to the high abundance of larvae above the beaver dam but was not included when calculating density, shown as an outlier in Figure 14.

Spawning Habitat Selection

Approximately 98% of wild-origin larvae emerged in the Gold Creek Pond outlet channel, indicating that there are strong influencing factors determining the location of adult spawners. Out of the five larvae that were recorded emerging in Gold Creek, only one larva was caught in Gold Creek above the confluence of Gold Creek and the Gold Creek Pond outlet channel. The four larvae captured downstream of the I-90 bridges in Gold Creek could have originally been from that specific reach of Gold Creek, but one cannot exclude the possibility that the larvae emerged in the outlet channel and missed drift nets until being captured farther downstream on their way to Keechelus Lake. Fall

2018 spawning surveys resulted in adults being observed residing in Gold Creek with evidence of redds, meaning that a lack of emerged larvae from Gold Creek was not a result of spawning adults completely avoiding Gold Creek altogether. However, the number of spawning adults and redds were higher in the outlet channel, with redds intermittently spanning the complete length of the channel until 100 m or so from the opening to Gold Creek Pond.

One apparent reason for the distinct lack of successful emergence of larvae in Gold Creek is the chronic dewatering problem Gold Creek experiences each year. As mentioned before, the drainage of Gold Creek first became altered due to the construction of Keechelus Lake dam in 1917, inundating the mouth of Gold Creek's drainage basin during seasonal high lake levels (Deichl et al. 2011; Natural Systems Design 2013). In the 1970s after the construction of Gold Creek Pond, severe dewatering of Gold Creek began, with the pond acting as a siphon drawing water away from Gold Creek (Kittitas Conservation Trust 2019). In lower gradient areas of Gold Creek, particularly in sections due west of Gold Creek Pond, portions of Gold Creek flow underground in summer months (Deichl et al. 2011). In addition, personal observations in the winter and early spring of 2019 showed that sections of Gold Creek began temporarily flowing underground in mid-March (Figure 25). These sections of dry gravel beds thus lead to the deaths of occupied redds that may have been laid there in the fall.

Figure 25. Photographs documenting early spring 2019 Gold Creek channel dewatering a) looking downstream and b) upstream on March 14th, c) and d) looking downstream and e) upstream on March 18th, f) looking downstream and g) upstream on April 7th.

Chronic dewatering of Gold Creek is not a new problem, but it has severe consequences to the health of the ecosystem. Documentation of the dewatering of Gold Creek has been going on for decades, with the observation that salmonids, particularly bull trout, becoming entrapped in pools. The majority of habitats chosen for redd placement by spawning bull trout in Gold Creek occurred after trout were isolated upstream by downstream channel dewatering (Wissmar and Craig 2004). Chronic seasonal dewatering has become such a pressing issue that actions are taken each year to rescue bull trout trapped in isolated pools (Mid-Columbia Fisheries Enhancement Group n.d.). The relatively short occupation time of spawning kokanee salmon in Gold Creek has no effect on bull trout distribution but there could be competitive interaction for foraging habitat between juvenile kokanee salmon and bull trout (Wissmar and Craig 2004). There is also a lack of scientific studies regarding the potential impact early spring Gold Creek dewatering has on fish distribution. Further site-specific studies are needed to address this topic in more depth.

An additional reason the majority of kokanee salmon spawn in the Gold Creek Pond outlet channel instead of Gold Creek could be the significant difference in water temperature between Gold Creek and the Gold Creek Pond outlet channel for the months of November 2018 through January 2019. On average the Gold Creek Pond outlet channel was 2 °C warmer than Gold Creek, possibly due to the fact that the outlet channel flows from Gold Creek Pond which is supplied by groundwater upwelling, as well as siphoning water from Gold Creek underground (Deichl et al. 2011). In contrast, Gold Creek is mainly fed by colder melting snow and ice upstream. Since only a single larva was captured in Gold Creek proper, upstream of the confluence of Gold Creek and the outlet channel, I was unable to compare if the emergence timing of larvae was different in the outlet channel versus Gold Creek as a result of differences in water temperature. For future studies following this line of questioning, I would recommend additional drift nets that can withstand velocities greater than 0.8 m/s. This is the maximum velocity our drift nets could withstand before being ripped out of the substrate, not counting the fact that mid-channel drift nets are most likely not feasible due to the high water velocity and the scoured hard substrate of Gold Creek during mid-spring when larvae emerge.

Early Life History of Kokanee Salmon

Characteristics and early life histories of hatchery-origin and wild-origin kokanee salmon differed in many respects. Hatchery-origin kokanee salmon sourced from the Naches Fish Hatchery were acquired as eggs from Lake Whatcom in mid-December. Hatchery kokanee salmon started an external feeding regime on January 30th, 2019, with water temperatures remaining between 8.9 °C to 10 °C. The hatchery-origin kokanee salmon were consistently inundated with food, even before full yolk absorption (Naches Fish Hatchery manager Matt Mathes, personal communications). Comparing hatchery natal environments to their wild counterparts, Gold Creek wild-origin kokanee salmon experienced greater fluctuations in water temperatures and food availability. These differences in rearing environments are reflected in otolith growth patterns.

Otolith microstructure differed between wild and hatchery-origin larvae, with hatchery-origin larvae having significantly larger distances between the central primordia and the exogenous feeding check, as well as between the central primordia and the exogenous feeding check. One can then infer that larger distances between checks is a result of wider daily growth increments. This difference in daily growth increments is supported by findings of a 2007 study by Barnett-Johnson et al., which concluded that daily growth increments in hatchery-reared Chinook were wider and more uniform immediately after the onset of the exogenous feeding check. Differences in food availability and consistency between hatchery and wild rearing environments can affect growth rates of otolith formation, resulting in differences in widths and patterns of daily growth increments (Barnett-Johnson et al. 2007). Though the Barnett-Johnson et al. study did not measure widths of daily growth increments before the exogenous feeding check, the fact that Naches Fish Hatchery kokanee salmon were inundated with an external food source before full yolk absorption, unlike wild-origin kokanee salmon, is a likely reason for increased daily growth increments before the exogenous feeding check.

Measurements between the central primordia and hatch check of wild and hatchery-origin larvae resulted in significantly shorter distances in hatchery larvae compared to wild larvae. These differences could be a result of hatchery-origin larvae experiencing a shorter incubation time between egg formation and hatching, possibly due to more uniform water temperatures compared to wider temperature fluctuations in the Gold Creek ecosystem. Having large significant differences in larvae otolith microstructures between wild and hatchery rearing environments is useful in order to analyze early life histories of adult kokanee salmon and determine rearing environment origins from selected otolith landmarks.

Adult Kokanee Salmon Stock Discrimination

Results show that out of a sample of 60 adult kokanee salmon, 30 collected in 2013 and 30 collected in 2019, 83% of the adults most likely were wild reared and 17% most likely were hatchery reared. Adults from the Gold Creek region collected in 2013 had 23 identified as most likely being wild in rearing origin and seven most likely being hatchery in rearing origin. Adults from Gold Creek collected in mid-October of 2019 had 27 identified as most likely being wild in rearing origin and three most likely being hatchery in rearing origin. It should be noted that the stock discrimination results from the LDA model using known-origin larvae is not an optimal linear discriminant analysis, where normally a prior estimate of stock composition is required (Barnett-Johnson et al. 2007). Discriminant analyses normally perform poorly when stock discrimination markers are similar (Campana 2005). In this instance, however, using a LDA was appropriate as there was a strong division in stock discrimination markers. Overall, the LDA model used in the research merely suggests at the possibly that the majority of kokanee salmon that spawn in Gold Creek have wild-rearing-origins.

Stock discrimination markers in adult kokanee salmon differed only for the parameter “distance from central primordia to hatch check,” with the distance not being significantly different between hatchery and wild-origin adults, unlike what was seen with larvae. One possible reason for the discrepancy is the small sample size of hatchery assigned adults (n = 10) used for the statistical analysis compared to the larger sample size (n = 20) of larvae. Future studies should increase the sample size of adult kokanee salmon used in the linear discriminant analysis in order to prevent a small sample size of hatchery assigned adults.

Future studies should be conducted using kokanee salmon from the Naches Fish Hatchery that have been physically marked, such as using adipose fin clipping, coded wire tags, or otolith thermal marking, in order to validate the stock discrimination of Gold Creek adults. Currently the Naches Fish Hatchery does not use physical tags to mark their kokanee salmon, resulting in the necessity of using otolith microstructure analysis to identify rearing origin. I would recommend using otolith thermal marking as it is a popular method used in hatchery-raised salmonids to identifying rearing origins after recapture. Hatchery managers can implement short-term temperature fluctuations in the hatchery rearing environment to create distinctive structural marks on otoliths, with the possibility to distinguish between brood stock years (Volk et al. 1999). Otolith thermal marking is optimal because it can mark 100% of hatchery salmon, becoming crucial as the majority of marked kokanee salmon do not make it to their spawning life stage due to recreational fishing and predation (Hansen et al. 2017). I would also recommend using otolith thermal marking as it requires no specialized equipment to be implemented and causes not discernable harm to the fish (Volk et al. 1999). Quantifying the contribution of hatchery-origin kokanee salmon to the Gold Creek system can only be validated using such techniques.

Gold Creek Management Implications

The Gold Creek ecosystem has a fraught history of natural resource exploitation, anthropogenic-driven changes, and major impacts to local fish populations (Deichl et al. 2011; Hansen et al. 2017; Kittitas Conservation Trust 2019). Despite habitat transformations, including the inundation of the mouth of Gold Creek during seasonal high lake levels, the chronic dewatering of Gold Creek, and the constriction of Gold Creek caused by the presence of Forest Service Road #4832, native fish populations, including kokanee salmon and ESA-listed bull trout populations, continue to persevere. The future of these species, however, is full of uncertainties.

The Forest Service, Washington Department of Fish and Wildlife, Yakama Nation Fisheries program, and other partners are currently developing plans to enhance fish and wildlife habitat and to reestablish habitat connectivity in the Gold Creek watershed. Goals for habitat restoration include integrating large woody debris and log jams into Gold Creek, increasing native vegetation along banks, increasing pools and side channels, and strengthening wildlife connectivity between habitats. Restoration efforts will include filling in Gold Creek Pond, removing the outlet channel, and transforming the land into a natural wetland habitat (Garvey-Darda and Kelly n.d.). However, as long as Gold Creek Pond continues to siphon water away from Gold Creek, leading to chronic dewatering that creates unsuitable habitat for salmonid spawning, continuing to have the Gold Creek Pond outlet channel as kokanee salmon spawning and rearing habitat will be critically important.

Results from this study show that there is a thriving kokanee salmon life cycle happening in the man-made Gold Creek Pond outlet channel, with the majority of adults most likely being wild in rearing origin. Future restoration efforts will need to integrate this information into their construction plans. Ultimately the removal of Gold Creek Pond and subsequent restoration of the region has the potential to allow Gold Creek to become a more suitable habitat to host kokanee salmon spawning and rearing.

If the majority of kokanee salmon that spawning in Gold Creek are wild in origin, that means that a large portion of the stocked kokanee salmon from the Naches Fish Hatchery are not contributing to the spawning in Gold Creek. The stocking of kokanee salmon in Keechelus Lake is probably mostly contributing to recreation fishing in the area, but also could be providing an essential food source for local bull trout. This emphasized the unique importance the Naches Fish Hatchery operation has on the Keechelus Lake and Gold Creek ecosystem. Additional studies are recommended to quantify the percentage of stocked kokanee salmon that are being consumed by bull trout and other piscivorous fish species, compared to recreational fishing purposes and the hatchery-origin adults that spawn in Gold Creek.

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