Fishery Data Series No. YY-XX

Evaluation of coho salmon aging methods and scale growth patterns using known age specimens

by

Kent F Crabtree

April 2015

Alaska Department of Fish and Game Division of Commercial Fisheries

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**Weights and measures (metric)**

centimeter cm

deciliter dL

gram g

hectare ha

kilogram kg

kilometer km

liter L

meter m

milliliter mL

millimeter mm

**Weights and measures (English)**

cubic feet per second ft3/s

foot ft

gallon gal

inch in

mile mi

nautical mile nmi

ounce oz

pound lb

quart qt

yard yd

**Time and temperature**

day d

degrees Celsius °C

degrees Fahrenheit °F

degrees kelvin K

hour h

minute min

second s

**Physics and chemistry**

all atomic symbols

alternating current AC

ampere A

calorie cal

direct current DC

hertz Hz

horsepower hp

hydrogen ion activity pH

(negative log of)

parts per million ppm

parts per thousand ppt,

‰

volts V

watts W

**General**

Alaska Administrative

Code AAC

all commonly accepted

abbreviations e.g., Mr., Mrs., AM, PM, etc.

all commonly accepted

professional titles e.g., Dr., Ph.D.,

R.N., etc.

at @

compass directions:

east E

north N

south S

west W

copyright ©

corporate suffixes:

Company Co.

Corporation Corp.

Incorporated Inc.

Limited Ltd.

District of Columbia D.C.

et alii (and others) et al.

et cetera (and so forth) etc.

exempli gratia

(for example) e.g.

Federal Information

Code FIC

id est (that is) i.e.

latitude or longitude lat. or long.

monetary symbols

(U.S.) $, ¢

months (tables and

figures): first three

letters Jan,...,Dec

registered trademark ®

trademark ™

United States

(adjective) U.S.

United States of

America (noun) USA

U.S.C. United States Code

U.S. state use two-letter abbreviations (e.g., AK, WA)

**Measures (fisheries)**

fork length FL

mideye-to-fork MEF

mideye-to-tail-fork METF

standard length SL

total length TL

**Mathematics, statistics**

*all standard mathematical*

*signs, symbols and*

*abbreviations*

alternate hypothesis HA

base of natural logarithm *e*

catch per unit effort CPUE

coefficient of variation CV

common test statistics (F, t, χ2, etc.)

confidence interval CI

correlation coefficient

(multiple) R

correlation coefficient

(simple) r

covariance cov

degree (angular ) °

degrees of freedom df

expected value *E*

greater than >

greater than or equal to ≥

harvest per unit effort HPUE

less than <

less than or equal to ≤

logarithm (natural) ln

logarithm (base 10) log

logarithm (specify base) log2, etc.

minute (angular) '

not significant NS

null hypothesis HO

percent %

probability P

probability of a type I error

(rejection of the null

hypothesis when true) α

probability of a type II error

(acceptance of the null

hypothesis when false) β

second (angular) "

standard deviation SD

standard error SE

variance

population Var

sample var

Fishery data report no. YY-XX

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Kent F Crabtree

Commercial Fisheries Division of ADFG, Douglas AK

Alaska Department of Fish and Game  
Division of Commercial Fisheries  
P.O. Box 240020 Douglas, Alaska, 99824

April 2015

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Kent F. Crabtree

Alaska Department of Fish and Game, Division of Commercial Fisheries

P.O. Box 240020, Douglas, Alaska 99824

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# abstract

Key Words: Coho salmon, Onchorhincus Kisutch, age, aging, scale pattern, growth, circuli, annulus, check, false-check, residual.

Accurate age determination is essential to the development and refinement of stock-recruitment relationships and the derivation of appropriate escapement goals (Beamish and McFarlane 1983). Coho salmon are commonly aged by interpretation of the growth pattern observed on the scale. In order to investigate the growth pattern and the ageability of coho salmon scales and their growth characteristics, known-age groups have been created and allowed to rear in natural native environments**.**

This study affirms that the accuracy of coho salmon age determination has been imperfect. It is evident that coho salmon scale patterns are often highly variable from system to system and from year to year as these patterns are influenced by the habitat types and the environmental conditons during the particular years from which they originate. This quality makes it very difficult to define rigid criteria for identification of legitimate winter annuli that may be applied to all coho salmon. A collection of known age smolt scale samples from different locations and over several years provides the ability to evaluate the variability of scale patterns between locations and between years, and provides an aging standard for the years of the study. The collection also provides an important training tool for those wanting to improve their ability to interpret coho salmon scale patterns.

A collection of scale pattern images of known age was produced by this study. It is an extremely valuable training tool for those seeking to improve their ability to interpret coho salmon scale patterns. In some situations, such as attempting to age coho from mixed stock fisheries where the nature of the habitat of origin is unknown, it may be concluded that coho scale patterns are variable to such a degree that a high level of accuracy is unobtainable. For particular stocks, reared in certain habitat types, under certain environmental conditions (that vary from year to year) perhaps the only means of acquiring highly accurate age determination is to produce a known-age group as an aging standard for every system and every year.

# introduction

Coho salmon rear in their natal freshwater habitats for one to three years. The marine life history is less variable. A large coho salmon spends approximately 16 months in the marine environment before returning to spawn; from the spring of a given year until the fall of the next which includes one winter in marine waters. There is a smaller component of any given cohort that return as jacks. These are individuals that spend only about four months in the marine environment; spring to fall of the same year, thus a winter is not included in their marine life history. Determining the age of coho salmon is primarily a problem of determining the age at smolting; the freshwater age. The marine age is usually easily established by size of the fish, size of the scale, and by the presence or absence of a winter annulus on the scale pattern. Though coho salmon that have lived in marine waters through two winters have been reported they are extraordinarily rare.

Coho salmon scale aging has been performed for many years as a component of stock recruit analysis and to further the understanding of the freshwater life history of juvenile coho salmon. Among those who age salmon scales coho salmon have developed a reputation for being among the more difficult. Our years of experience examining coho scale patterns has shown that the freshwater rearing portion is not very stable from year to year and that certain predictable characteristics are associated with different systems or rearing locations. In particular two of our study areas are the Berner’s River and Hugh Smith Lake from which we have collected smolt and adult scale data for over 30 years. This includes a seventeen year period of conducting an age validation study. The scale samples from these two locations predictably display distinctly differing traits from each other. The most obvious differences are in the concentration of circuli and the relative “strength” of the annulus.

Fish scales develop by adding incremental rings of growth that are discernable to the eye as concentric circles. Each of these incremental rings is termed a circulus. The spacing of the circuli are generally thought to be an indication of the rate of growth. A group of tightly spaced circuli is termed a “check”. In conjunction with the onset of Winter conditions a group of circuli with reduced spacing results and typically there are also intermittent breaks in one or more circuli; this is the winter check which in this report will be called a “winter annulus” or more simply an “annulus” since it appropriately indicates the passage of a year from the previous winter. Checks similar to the annulus but unrelated to winter conditions and timing can also occur. These other checks are the major impediment to accurate aging especially when found among patterns bearing weak but legitimate winter annuli.

I refer to “lake patterns” versus “river patterns” as a result of the observations made regarding the samples from the system types included in this study. The lake pattern displays a higher concentration of circuli, the circuli being more numerous and packed more tightly together. Also the winter annuli are often weakly manifested and the circuli spacing tends to lack a high degree of seasonal graduations. The river pattern is characterized by fewer total circuli whith wider spacing. The winter annuli are usually more strongly manifested and the circuli spacing is frequently well graduated becoming narrower nearer to the winter annulus. Checks can occur, to varying degrees, with either of these pattern types.

The extant literature concerning coho salmon aging offers nothing for the validation of ages or aging techniques applied to fish reared in the wild in their native habitats. The few studies done concerning scale growth and aging are relatively short-term investigations that include a single winter at best and are always performed in artificial environments such as a laboratory or a hatchery. This literature is sparse and inconclusive in terms of offering knowledge on how to interpret a scale pattern from a wild specimen reared in their natural environment over the duration of their freshwater life history. [find study that suggested a rapid increase in growth may have caused a check]

Due to the lack of adequate studies and our observations of these differing pattern types, and also the notable differences in the pattern characteristics from year to year for the same system it was necessary to undertake this investigation. This study evaluates coho scale patterns in detail to improve our understanding of scale features and their range of variability with the hope of achieving improved aging accuracy.

[Where is this comment applicable? Include comment regarding Auke lake?]

**Methods**

This study creates a sub-population of wild rearing coho salmon that are coded-wire tagged as fry that have emerged in recent days or weeks such that their age when recaptured and sampled at a later stage in life can be determined. The scale patterns from these samples are then examined to determine how they relate to the age of the fish. Two systems were chosen for study in order to compare the scale patterns from different habitat types: Berners River as a “river type” habitat (characterised by shallow waters and various wetlands.) and Hugh Smith Lake as a “lake type” habitat. Methods used to capture both fry and smolt differed at the two systems as a result of these habitat differences.

This study was greatly facilitated by the fact that long-term, coho salmon stock assessment studies were already established (in the early 1980s; Shaul et al. 2011) and conducted annually at both Berners River and Hugh Smith Lake. These stock assessment studies included capturing, sampling, and coded-wire tagging smolt in the spring, and capturing and sampling adults in the fall. Thus, infrastructure, experienced field crews, equipment, and logistical support were already available for these remote field sites. The only additional procedures required for a known-age study were the capture and tagging of newly emerged fry and the recovery and sampling of known-age specimens concurrent with ongoing smolt trapping and sampling activities. Known-age studies were initiated at Berners River and Hugh Smith Lake in 1996 and 1997 respectively. Known-age studies were also conducted on coho salmon by Craig Farrington at Auke Lake from 1992 to 1997, and some of the results and samples are incorporated into this report.

**Study Sites**

**Hugh Smith Lake**

Hugh Smith Lake is located 97 km southeast of Ketchikan on the Southeast Alaska mainland in Misty Fjords National Monument (55˚ 06’ N, 134 ˚ 40’ W; FIGURE?). The lake is organically stained, with a surface area of 320 ha, mean depth of 70 m, and maximum depth of 121 m. It is meromictic, and water located below 60 m does not interact with the upper freshwater layer of the lake. The lake drains into Boca de Quadra inlet via 50-m long Sockeye Creek and is supplied by two major inlet streams: Buschmann Creek flows northwest 4 km to the head of the lake and Cobb Creek, the primary spawning stream, flows north 8 km to the southeast head of the lake. Cobb Creek has a barrier to anadromous migration approximately 0.8 km upstream from the lake.

Coho salmon rear in both the inlet streams and around the shoreline of the lake. Much of the shoreline is relatively steep and rocky, with limited vegetation; however, a large log jam at the outlet of the lake and numerous deadfalls and rock slides around the shoreline provide important habitat structure for rearing juveniles (Shaul et al. 2009). Coho salmon production from Hugh Smith Lake is relatively low for the amount of surface area and the length of stream and shoreline area compared to other Southeast Alaska lake systems (Shaul et al. 1985; Shaul and Van Alen 2001).

**Berners River**

The Berners River is located 69 km northwest of Juneau (58˚ 23’ N, 134˚ 38’ W; FIGURE?). It flows southeast 24 km into upper Berners Bay, and is a relatively compact drainage. The major spawning ground is the headwater area upstream of the confluence of two river branches and above the glacial tributaries. There are minor spawning locations in the lake that connects to the east branch and in other small tributary streams (Gray and Marriott 1986). Most coho fry migrate downriver several kilometers to rear in a expanse of prime habitats mostly consisting of relatively shallow water wetlands including; beaver ponds, sloughs and marshes. [[ More needed ?? ]]

**Auke Lake**

Auke Lake is located 19 km northwest of Juneau (58˚ 23’ N, 134˚ 38’ W; FIGURE?). The lake has a surface area of 67 ha, mean depth of 19 m, and maximum depth of 31 m. The outlet stream, Auke Creek, is less than 1 km long and of moderate gradient, with few natural pools or spawning gravel except for a small number of man-made backwater pools filled with gravel for spawning. The main inlet stream, Lake Creek, is the largest of five tributary streams to the lake, and is a low gradient stream of pools and riffles with an abundance of suitable spawning gravel. Much of the lake is bordered by forest; however, at least 50% of the shoreline has been urbanized by residential development (Lum and Taylor 2004). The lake shore includes areas dominated by emergent vegetation, such as *Equisetum* sp. and *Nuphar* sp., while other areas are characterized by large quantities of woody debris (Lum and Taylor 2004).

The NOAA Fisheries, Alaska Fisheries Science Center, Auke Bay Laboratories, operates the Auke Creek Research Station located on the outlet of Auke Lake in conjunction with ADF&G and the University of Alaska, Fairbanks. The research station maintains a concrete and steel fish weir structure located on Auke Creek just above the highest high tide level (Lum et al. 1998). The weir was operated annually in a downstream trapping configuration from early March through June to intercept emigrating salmon smolt and trout, then changed to an upstream trapping configuration and operated through early November to capture migrating adult salmon. It is assumed that 100% of migrating salmonids were captured in the weir trap.

**Hugh Smith and Berners Studies**

**Fry Capture and Tagging**

Annually a target of 5,000 fry were coded-wire tagged at each site anticipating a yield of 100 to 300 potential known-age smolt samples each year. A maximum size limit of 42 mm (snout to fork length) was initially established to ensure that the tagged fry were indeed young-of-the-year. This initial size limit was determined from a small quantity of length frequency data collected at Berners River in 1996; however, later field observations suggested that some fish in the low and sub-40 mm range may not be newly emerged fry. Fish of this size were occasionally seen having the coloration and subtle body shape characteristics of age-1 juveniles. Also, juvenile coho salmon less than 40 mm were captured during November from tributaries to Hugh Smith Lake; since little or no growth is thought to occur during the winter months, this suggests that very small fish may be found in the spring that are not the progeny of the most recent brood year, ie. not recently emerged fry. As a result of these findings, the maximum size was reduced to 38 mm. Half-length coded wire tags were used to tag these tiny fish. [[ Insert info on tagging methods? – or could say methods described by Magnus et al. 2006? ]]

Coho fry were captured annually at Hugh Smith Lake from approximately mid-May to early June using a fyke net (1 m tall × 2 m wide at the mouth) installed near the mouth of Cobb Creek, the primary spawning stream. Newly hatched fry were passively captured as they migrated downstream and were funneled into the fyke net. A lidded holding box at the terminal end of the net protected captured fry from the stream current and predators. Fry were collected daily and transported in buckets to the tagging site at the lake outlet where they were adipose-clipped, coded-wire tagged, and held overnight in holding pens to check for tag retention and mortality. Tagged fry were transported back to Cobb Creek and released downstream of the fyke net to avoid recapture.

At the Berners River, coho fry were captured annually from approximately mid-May to early June. Newly hatched fry congregated in eddies and small sloughs out of the main river flow after drifting downriver several kilometers from the spawning areas. They were also concentrated in good numbers in quiet water where spills from beaver ponds occurred. Presumably they were positioned to gain access to the beaver pond areas, preferred rearing habitat, during high water events. Fry were simply captured by stalking these holding areas and scooping them up with a 1/8” or 3/16” mesh dip net. Typically 100–400 fry were tagged per day, a quantity that could be captured in 10–15 minutes of dip netting effort. Captured fry were transported in buckets to the field tagging station at the main camp where they were adipose-clipped, coded-wire tagged, and held overnight to check for tag retention and mortality. Tagged fry were again transported in buckets and released into a large beaver pond complex (Shaul Pond) where most of the fry had been captured near the base of outspills.

Known-Age Smolt Collection

Coho smolt were captured each year from mid-April to early June at a weir operated at the outlet of Hugh Smith Lake. The weir, composed of plastic screen panels supported by a cable across the stream and an incline plane trap to capture fish, was described in detail by Magnus et al. (2006) and Shaul et al. (2009). Coho smolt were captured annually at the Berners River from approximately early May to mid-June using two styles of traps (Shaul and Crabtree 2014). Custom-built traps, similar to oversized Gee minnow traps (Magnus et al. 2006), were baited with salmon roe and fished in the slough and river, and passive “spill traps” were installed in beaver ponds. Spill traps consisted of a dewatering trough placed in the effluent flow from a beaver pond—water was directed through a flexible 4 in. pipe attached to the end of the trough and into a floating holding box where migrant smolt were captured. A large perforated panel on one side of the box allowed for water flow. Other spills along the beaver dam were blocked with sand bags or plastic mesh to concentrate flow at the trough. Four or five of these traps were installed at different locations in the drainage. (Photographs of various Berners River spill traps were provided by Magnus et al. 2006, pages 60–62).

As captured smolt were processed for coded-wire tagging, those fish already missing their adipose fin were set aside as potential known-age specimens. Adipose-clipped smolt were passed through a coded wire tag detector to determine which specimens contained tags. Those with tags were carefully examined at the adipose excision to determine if the cut was fresh or healed over. Only those that were healed were collected as known-age samples. A few samples turned out to be fish that were tagged as presumed smolt the previous spring but which remained in freshwater another year. This error occurred almost exclusively with smolt captured at the Berners River in the slough rather than beaver pond areas, since those areas were semi-isolated (flood events provide opportunity for pond access) from one another and fish tagged as smolt were released into the slough.

Smolts that were selected as known-age samples were measured (snout to fork length, mm), weighed (to the nearest 0.1 g), and sampled for scales. Ten to fifteen scales were taken from the left side of the fish approximately two rows above the lateral line along a diagonal downward from the posterior insertion of the dorsal fin to the anterior insertion of the anal fin (INPFC 1963). The scales were collected by scraping with a surgical scalpel and then were spread on a glass slide, two samples per slide. An individually numbered Floy tag was attached to the smolt carcass and the entire fish was preserved in a bottle of ethyl alcohol. These samples were later delivered to the ADF&G Mark, Tag and Age Laboratory where coded wire tags were removed and decoded. Using this tag information ages are assigned to the collected samples.

**Auke Lake Study**

As a routine part of monitoring the Auke Lake coho salmon population, all smolts and adults are counted as they pass the weir, and examined for presence or absence of a coded wire tag. Additionally, before release downstream of the weir all emigrating coho smolts were sorted into four size categories by fork length: small (<90 mm), medium (90–110 mm), large (111–125 mm), and extra-large (>125 mm).

In October 1992, 13 adult female and 11 adult male coho salmon were captured at the weir, held for several weeks, and artificially spawned in November, which is similar to the natural spawning time in the Auke Lake system. The fertilized eggs were incubated in Heath trays at the Auke Creek Research Station. Water for incubation came from Auke Creek, and matched water temperature regimes expected for naturally spawned coho salmon eggs. A total of 10,411 of the resulting coho fry were marked with an adipose clip. During 25–28 May 1993, they were planted as unfed fry into two different habitat types in the Auke Lake watershed: 1) shallow, weedy waters along the lake margin, considered prime juvenile coho salmon rearing habitat, and 2) pools in Lake Creek where coho fry were frequently observed in spring. Adipose-clipped fish were recovered as migrant smolt at the weir during April–June in 1994 and 1995. Not all marked fish were sampled. In 1994, only marked fish up to 110 mm fork length were sacrificed for known-age samples, 125 of 488 mark recoveries. In 1995, all mark recoveries, 61 fish total, were sacrificed for known-age samples.

In spring 1995, nomadic, young-of-the-year wild coho fry were captured in the downstream trap of the weir. Additional fry were caught with dip nets upstream of the weir to supplement weir numbers. Fry were measured (snout to fork length, mm), marked with an adipose clip, and tagged with half-length coded wire tags in groups according to three release sites: Lake Creek, Auke Lake, and Auke Creek. Each group was sampled for length (snout to fork, mm) and absence of scales, and then released on 2 June, 19 June, and 19 June 1995, respectively. Pre-release tag retention was 100%, 100%, and 99.5%, respectively. In summer and fall 1995, baited minnow traps were used to catch juvenile coho salmon in Auke Lake. The few adipose-clipped fish that were captured were checked for the presence of a coded wire tag using a field detector, sampled for length and scales, then released back into the lake. Adipose-clipped fish were also recovered and sampled as migrant smolt at the weir during April–June 1996 and 1997, all of which were sacrificed for tag removal.

Auke Lake coho smolt were measured, weighed, and sampled for scales following the same protocols used at Hugh Smith Lake and the Berners River. [[ More on how samples were shared with ADF&G; other stuff? ]]

Age determinations were based on examinations of scale images at 70× on a microfiche viewing projector. Criteria used to assign ages were similar to those of Mosher (1968). Photographs of selected scales were taken on a Leitz Laborlux S (trademark) microscope mounted with a Wild Leitz MPS 46 Photoautomat (trademark) camera using 35 mm Kodak Kodalith (trademark) film.

# METHODS

The capture methods used at the two systems differ for both fry capture and smolt capture. At Hugh Smith Lake the fry are captured by using a fyke net installed in a tributary of the lake, Cobb Creek. This captures newly hatched fry as they drift with the current towards the lake. The mouth of the fyke net is about 1 M tall by 2 M wide. It has a holding box at the downstream terminus where the fry collect and find refuge from the current. -------The smolt are captured at the outlet of Hugh Smith Lake using a smolt weir installation with an inclined plane trap.

Smolt capture is achieved by using “spill traps”. These are a passive trap that requires a few inches of head or drop in water level to function. They are installed in the perimeter of beaver ponds where there is effluent flow. A dewatering trough is installed to receive the effluent, it terminates with a 4 inch pipe that directs the remaining flow into a floating holding box. As migrating smolt follow the flow out of the beaver pond they pass through the trough and the connecting pipe and are captured in the holding box. The box has a large perforated panel on one side that allows flow. Four or five of these traps are installed at different locations. They are checked once or twice a day depending on the run timing and intensity of smolt movements.

As the smolt catch is processed for coded wire tagging those that are missing the adipose fin are put aside as potential known age specimens. When the days tagging is completed the adipose clipped smolt that have been discovered are run through the coded wire tag detector to determine which specimens contain tags. Those with tags are carefully examined at the adipose excision to determine if it is a fresh cut or if the cut is healed over. Only those that are healed are collected as known age samples. A few samples turn out to be fish that were tagged as presumed smolt the previous spring but which remained in freshwater another year. This error occurs almost exclusively with smolt captured in the slough versus the beaver pond since those areas are semi isolated from one another and tagged fish as smolt are released into the slough. The smolt that are selected are sampled for scales and length. An individually numbered floy tag is attached to the smolt and the entire fish is preserved in a bottle of ethyl alcohol. ------

# Methods used in Auke Lake study

Study site

The Alaska Department of Fish and Game in cooperation with the National Marine Fisheries Service Auke Bay operates the Auke Creek Research Station located on the outlet from Auke Lake. Auke Lake is a productive, moderately deep lake covering 92 hectares with a heavily forested shoreline. The lake and its tributaries support indigenous populations of coho, sockeye, and pink salmon, cutthroat trout and dolly varden char, as well as incidental numbers of chum salmon and steelhead trout. The outlet stream, Auke Creek, is less than 1 kilometer long, of moderate gradient with few natural pools or spawning gravel except for a small number of man-made backwater pools filled with gravel for spawning. The main tributary to Auke Lake, Lake Creek, is a low gradient stream of pools and riffles with an abundance of gravel suitable for adult coho spawning. Auke Creek Research Station maintains a concrete and steel fish weir structure located on Auke Creek just above the highest high tide level. The weir is operated in a downstream trapping configuration from early March through June to intercept emigrating salmon smolt and trout, then changed to the upstream trapping configuration and operated through early November for migrating adult salmon. It is assumed 100% of migrating salmonids are captured in the weir trap.

Fish enumeration and aging

As a routine part of monitoring the Auke Lake coho salmon population, all smolts and adults are counted as they pass the weir, and examined for presence or absence of a coded-wire tag. Additionally, before release downstream of the weir all emigrating coho smolts were sorted into four categories by fork length size: small - less than 90mm, medium - 90-110mm, large - 111-125mm, and extra large - greater than 125mm.

Smolt scales are taken in a systematic fashion to include all size categories. Scales are taken from the preferred area (INPFC 1963), and mounted on glass slides. Age determinations were based on examinations of scale images at 70X on a microfiche viewing projector. Criteria used to assign ages were similar to those of Mosher (1968). Photographs of selected scales were taken on a Leitz Laborlux S (trademark) microscope mounted with a Wild Leitz MPS 46 Photoautomat (trademark) camera using 35mm Kodak Kodalith (trademark) film.

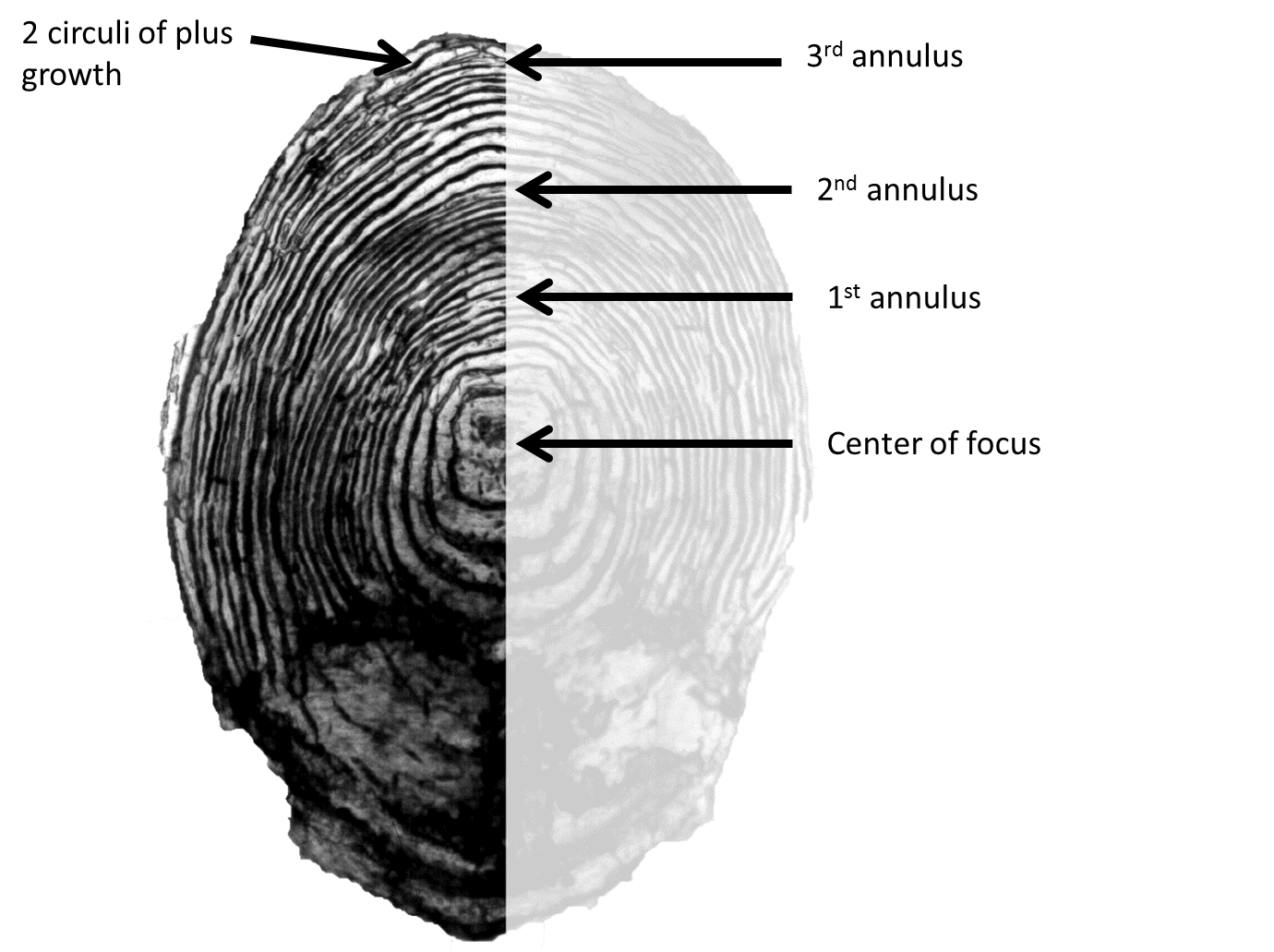
Marking and tagging

In October 1992, 13 adult female cohos and 11 adult male cohos migrating to Auke Lake were captured at the weir and held for several weeks. They were artificially spawned in November, which is timing similar to natural coho spawning in the Auke Lake system. The fertilized eggs were incubated in Heath (trademark) trays at the Auke Creek Research Station. Water for incubation came from Auke Creek, matching water temperature regimes expected for naturally spawned coho eggs. A total of 10,411 of the resulting coho fry were marked with a fin clip and planted as unfed fry into the Auke Lake watershed from May 25 - 28, 1993. The plantings were made into two different habitat types: into shallow, weedy waters on the lake margin, considered prime juvenile coho rearing habitat; and into pools in Lake Creek where coho fry are frequently seen in Spring.

Recoveries of marked smolts were made at the weir during daily sorts from April through June, 1994 and 1995. Not all marked fish were sampled. In 1994, only marked fish up to 110mm fork-length were sacrificed for samples, 125 out of 488 mark recoveries. In 1995, all mark recoveries, 61 fish in total, were sacrificed for samples.

In Spring 1995, nomadic, young-of-the-year wild coho fry were captured in the downstream trap of the weir. Additional fry were caught with dip nets upstream of the weir to supplement weir numbers. Fry were marked with a fin clip and tagged with half-length coded-wire in groups according to three release sites, Lake Creek, Auke Lake, and Auke Creek. Each group was sampled for fork-lengths and absence of scales, and then released on June 2, June 19, and June 19, 1995, respectively. Pre-release tag retention ran 100%, 100%, and 99.5%, respectively. In Summer and Fall 1995, baited minnow traps were used in Auke Lake to catch tagged juvenile cohos. The few marked fish captured were checked for presence of a coded-wire tag using a field detector, sampled for fork-length and scales, and then released back into the lake. Recoveries of marked and tagged smolts were made at the weir during daily sorts in April through June 1996 and 1997. All were sacrificed for tag removal.

THE SMOLT SCALE



## data production

These samples are later turned in to the ADF&G CWT lab where they are dissected to recover the tag and the tag code is used to determine the year that the specimen was tagged as a fry, thus the age of the smolt is established.

In preparation for technical analysis a scale from each sample is selected and a digital image is created. The scale images are then processed by an image analysis software program using Optimate. This process produces a set of incremental measurements from the center of the scale to the outer margin circuli by circuli. An operator designates the measurements as different zones corresponding to the year of growth. From the focus to the end of the first annulus all measurements are designated as zone 1, continuing from there to the end of the second annulus all measurements are designated as zone 2. Growth that is beyond the outermost annulus is referred to as “plus growth”. For a 1.0 age smolt, plus growth is designated as zone 2. For a 2.0 age smolt, plus growth is designated as zone 3.

The resulting data is then entered into a database format that is organized to have one row per circulus measurement. Using this database, descriptive statistics are derived that characterize various features of the scale patterns according to age.

# Discriminant analysis to classify coho smolt

**Data and Method**

Coho salmon smolt (age-1 or 2) were sampled from three stocks (AL, BR, and HS). AL stock were sampled in 1994-1998 and 2000; both BR and HS stocks were sampled in 1998-2005. Scales were collected for the scale pattern analysis (SPA). We were able to obtain up to 50 quantitative variables for each individual fish from SPA (Table 1). Among these 50 variables, some of them may not be retrieved from the age-1 smolt due to their younger age or slow growth. We excluded those variables from our analysis. The rest of variables (bold in Table 1) were applied on repeated iterations of STEPDISC (SAS, Inc.) to select the subset of variables that best differentiated the ages. This selection process was applied separately for each stock and selected subset of variables may differ in stocks. The subset of variables was applied to the discriminant analysis for each stock.

Discriminant analysis is used to classify an individual fish into either of two known age groups on the basis of a set of quantitative variables from SPA. The analysis first develops a discriminant criterion to classify each individual fish into one of the two age groups. The dataset used to derive the discriminant criterion is called the training or calibration data set. The derived discriminant criterion can be applied to a second data set (test data set) to assign individual fish into age groups. We assume that the distribution within each age group is multivariate normal so that a parametric method can be used to develop a discriminant function. The performance of a discriminant function can be evaluated by estimating error rates (probabilities of misclassification). Error count estimates and posterior probability error rate estimates can be evaluated in the analysis.

We developed discriminant criterions separately for each of three stocks. We used 1994-1998 data set as training data to develop a discriminant function for AL stock; 1998-2004 for BR and HS stocks. Then we applied the criterions to the latest year data set (test data set): 2000 data set for AL stock; 2005 data set for BR and HS stocks.

We also pooled the three stocks and used pooled 1994-2004 data set as training data to develop a discriminant function for all the stocks and the used 2005 pooled data as test data set for its evaluation.

# Observations of scale characteristics







For the lake systems both the scale size and the circuli number are correlated with the fish size, however the number of circuli has greater variability and a weaker correlation. Berner’s River exhibits greater variability for both scale size and number of circuli relative to fish size. Fig 1-6.(regression plots)



Smolt scale axis lenght is comparable for the lake systems. Both in breadth of distribution and in central tendency (mean, medan). Scale size ranges from approximately 350 to 750 microns for the age 1 smolts at these systems. Age 2 smolt size ranges from 450 to 800 microns with an average near 625 microns.

Berner’s River smolt scales tend to be slightly smaller than those from the lake systems. Age 1’s range from 300 to 650 microns withamedian of 450 microns. Age 2’s range from 350 to 750 microns with median of 540 microns. Figure 7-9. (3 charts of scale size)[ranges and medians used above are approximate, need refinement]







Common to all three systems the age 1 smolt possess the greatest number of circuli and the age 2 smolt, in their second season of growth (Z2), produce the fewest. Interestingly it is consistently seen that for the age 2 smolt the first growth season (Z1) has produced fewer circuli than the first growth season of their age 1 counterparts. This suggests the contingent that smolt at age 1 have been the more successful at growth.

As seen from these charts generally the lake system habitats, Hugh Smith Lake and Auke Lake, produce greater numbers of circuli in the first year of growth (Z1) than does the habitat of Berner’s river which consists of smaller volume and shallower water bodies such as stream channels, sloughs and beaver ponds. Auke Lake is much smaller than Hugh Smith Lake and it is located at a more northerly latitude and the Z1 growth is smaller.

For Auke Lake the Z1 and Z2 for age 2.0’s is roughly equivalent.

Only two examples of age 3.0’s have been obtained, both from Bener’s River. Surprisingly the circuli numbers for Z3 increase. Perhaps the fish at this age have reached a threshold size that allows them to feed more successfully or more competetively than the younger/smaller juveniles.

The difference between circuli counts for Z1 of age 1.0 and age 2.0 is present for the Berner’s River samples but is less significant that it is for the lake systems because there is overall a lower number of circuli presumably due to a shorter favorable season of growth.

For the majority of individuals Z1 has a greater number of circuli than does Z2 for both the Berner’s River and the Hugh Smith Lake Systems. Growth at Hugh Smith Lake is consistently greater than at Berners River as demonstrated by both circuli number and scale size.

Lake habitat maintains a more stable growth environment, particular regarding temperature, and provides a longer growth season.

# Investigation of non-winter check formation

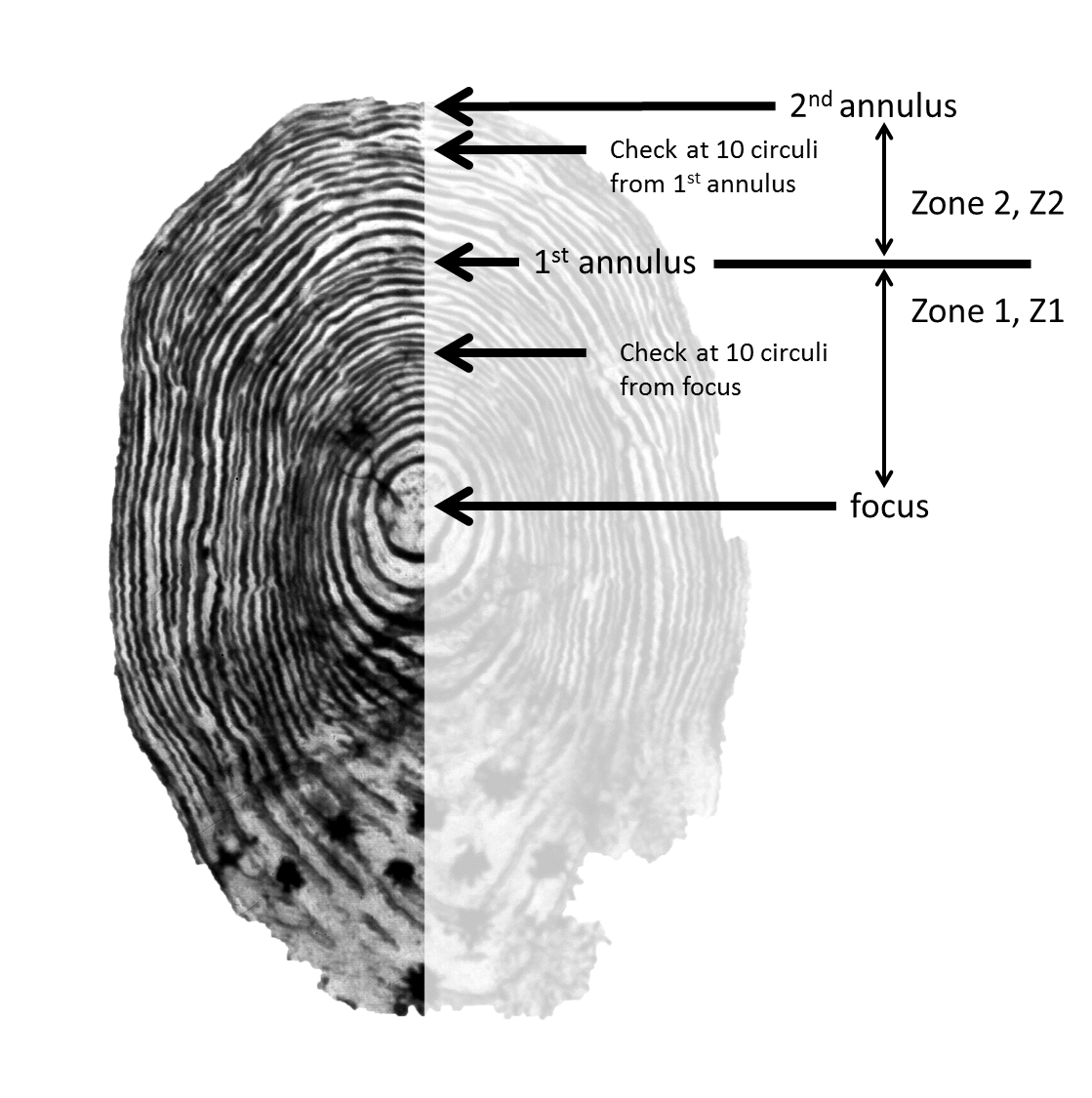
The problem of aging coho salmon stems largely from their propensity for check formation. Coho will form checks by varying degrees presumably depending on environmental conditions. Though infrequent it is not too unusual to find checks that are stronger or more significant in appearance than the annulus; the check that formed during the winter months. Variability in the characteristics of the annular check is also an impediment to proper aging. The annulus can involve approximately two to four circuli with graduated narrowing of circuli preceding the annulus or the graduated element may be muted or absent. The various characteristics that define the”annulus” are to a degree plastic and are best interpreted relative to a particular year and location.

To evaluate the phenomenon of check formation a subsample of 1,335 specimens from the known age collection was carefully examined. Checks were noted by strength and location. Strength was judged on a five degree scale from very strong to very-very weak. Check location was noted as occurring in either the FW1 or FW2 zone and by the circuli count from the focus. Samples that were judged likely to be misaged were also noted.

This analysis makes possible a number of observations regarding the frequency of check formation, where they are likely to form in the scale pattern and also the degree of variability that can be expected between locations and between years.

Sub-samples were extracted from the sample collections from Berner’s River and Hugh Smith Lake years 1998 through 2005 and from Auke Lake years 1998 and 2000 where comparison can be made between the same age fish for the same years. The sub-samples were selected to include a distribution of samples from throughout the sample period and throughout the size range but with weighted preference for those collected early and late in the sample period and for the larger specimens. The samples were selected without referring to their scale image but by considering only their sample date and snout-fork length.

**Check Locations**



Hugh Smith Lake smolt sample (#1039) from year 2000, age 2.0, 139mm.

In this image a check occurs in both of the freshwater growth zones, both Z1 and Z2, and they are each located at the 10th circuli of growth for the season.

|  |  |
| --- | --- |
|  |  |
|  |  |

The charts on the top include checks of all rankings. The bottom charts show only the stronger checks that were ranked 4 or 5 from the 5-point scale. The y-axis is the number of samples with a check at the location defined by the circuli count on the x-axis.

These charts show the location of checks by the circuli count from the focus, except in the case of checks found in the second year of growth, where the circuli count has been adjusted to be the circuli count from the start of the growth season. In this way the check location in Z2 can be compared to those in Z1 making them both relative to the beginning of the growing season for the year.

The top charts show the distribution of all checks that were noted at all strength rankings. These are informative for displaying the range where checks may develop. Regarding issues of aging the bottom charts show the location of only the checks that are prominent enough that they might be mistaken for an annulus.

For both Hugh Smith Lake and Berner’ River, whether the first or second year of growth, the location of checks, when they occur, are very similar. Checks occur most commonly between 6 to 9 circuli into the growth season. However for Berner’s River age 1’s a high rate of occurrence extends as far as the 12th circuli and for Hugh Smith Lake checks may be found at gradually diminishing frequency out to 20 circuli.

**The incidence of checks**

Checks occur at over twice the rate in the lake systems as compared to Berner’s River where the overall incidence of checks is 28.6%. The rate is 61.1% at Auke Lake and is 63.2% at Hugh Smith Lake. The greater number of checks results in a much higher rate of misaging. The rates of misaging for all years and all ages is only 1.2% for Berner’s River while it is 8.3% for Hugh Smith Lake and is highest at Auke Lake where it reaches 13.9%.

Checks that are mistaken for an annulus result in the error of overaging. This is far more common than underaging that can results when an annulus is so weak that it is mistaken for a non-annular check. Only one specimen was noted as likely to be under aged, an age 2 from Berner’s River while there were a total of eight samples from Berner’s River that were deemed likely to be over aged. Most of the over aging occurs among the age 1’s with the proportion varying between systems. At Berner’s River age 1’s could be overaged at three times the rate as age 2’s. For Hugh Smith Lake over aging of age 1’s occur at twice the rate of over aging age 2’s. Auke Lake yields five specimens that might be overaged, two age 1’s and three age 2’s, this is an inverted proportion compared to the other systems which is likely do to the small sample size from Auke Lake.

The estimates for misaging are quite variable from year to year and especially between system types. Berner’s River misaging rates range from zero to a high of 3.4% whereas at Hugh Smith Lake rates do not fall below 5% and range as high as 20%. The rates are similarly high at Auke Lake.

Check formation is highly variable; generally it occurs at a much higher rate at Hugh Smith Lake than at Berner’s River. For the age 1’s Hugh Smith Lake has nearly twice the incidence of check formation at 64% versus Berner’s River’s 38%. Similarly for age 2’s in the FW2 zone check formation occurs at twice the rate as at Hugh Smith Lake however in the FW1 zone the rate jumps to five times that of Berner’s River at 56.1% versus 11.5%.

This high rate of checks in the FW1 of Hugh Smith Lake age 2’s inspires speculation that the checks are indications of growth slow-downs. At Hugh Smith Lake the proportion of age 2 smolt is much smaller than at Berners river (insert stats), presumably these fish have failed to grow as rapidly as the majority of the cohort and have fallen short of reaching the size /fitness threshold required to successfully smolt at age one; perhaps the high check rate is an indication of that.

This data indicates that a high rate of checking does not necessarily result in a high rate of misaging. For Berner’s River the year with the highest overall incidence of checks (37.2%) resulted in a misaged rate of only 1.1%. The second to lowest incidence of check (19.5%) resulted in the highest misaged rate for Berner’s River of 3.4%. For Hugh Smith Lake the years 1999, 2001, and 2002 each had an overall incidence of checks of 70.0%. Those years had misage rates of 6.0%, 10.0%, and 20.0% respectively. The 20.0% rate was the highest observed in this study. The lowest rate of misaging at Hugh Smith Lake, 5.4%, occurred concurrent with an overall check incidence rate of 67.6%, which is slightly under the median rate of 70%.

Misaging as overaging may result when a check occurs that is sufficiently strong to be mistaken for an annulus and when the spacing between this check and the true annulus is enough to be mistaken for a summer season of growth. Misaging as underaging may occur when an annulus is not recognized. This can result when there is a lack of graduated spacing near to the annular check and when only two or perhaps three circuli appear to comprise the annular check.

A subsample from the collection of known age samples was inspected to generate the table X.X.

Scales were examined for the presence of growth disturbances in the pattern of circuli that would be considered a “check”. If a check occurred its location was recorded as the circuli count from the focus to the center of its location.

This investigation yields information on the frequency of occurrence of checks and their location on the scale by year and system. The strength of checks was also noted.

Berners River; for all years and all ages the overall rate of check formation is 28.6%. For all years the incidence of checks among age1 and age 2 is 38.0% and 21.6% respectively. For the age 2’s check occurrence is similar between FW1 and FW2 at 11.5% and 11.0% respectively.

Hugh Smith Lake; for all years and all ages the overall rate of check formation is 63.2%. For all years the incidence of checks among age 1 and age 2 is nearly equivalent at 64.2% and 61.4% respectively. For the age 2’s check occurrence is more than 2.5 times as frequent in FW1 than in FW2 at 56.1% and 21.2% respectively.

For all the years it is seen that the lake system (Hugh Smith Lake) has a much higher incidence of check formation at over twice the rate, 63% versus the stream/beaver pond system (Berner’s River) at 29%. It is also seen that for age 2’s the incidence of check formation is about the same for FW1 and FW2 at Berner’s River (11.5%, 11.0% respectively), however at Hugh Smith Lake the checks are formed in FW1 at nearly three times the rate as in FW2 (56.1%, 21.2% respectively).



|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  |  | Sample size | |  |  | False Check occurence | | | not a sum | overall | incidence | incidence |  |  |
| **Site** | **Year** | **Age 1** | **Age2** | **Age 3** | **total** | **age 1** | **age2 FW1** | **age2 FW2** | **age2 total** | **incidence** | **Age 1** | **Age 2** | **FW1** | **FW2** |
| BR | 1998 | 33 | 57 | 0 | 90 | 13 | 11 | 0 | 11 | 26.7 | 39.4 | 19.3 | 19.3 | 0.0 |
| BR | 1999 | 45 | 55 | 1 | 101 | 15 | 5 | 4 | 8 | 22.8 | 33.3 | 14.5 | 9.1 | 7.3 |
| BR | 2000 | 43 | 37 | 0 | 80 | 10 | 2 | 0 | 2 | 15.0 | 23.3 | 5.4 | 5.4 | 0.0 |
| BR | 2001 | 50 | 37 | 0 | 87 | 15 | 1 | 2 | 2 | 19.5 | 30.0 | 5.4 | 2.7 | 5.4 |
| BR | 2002 | 23 | 59 | 1 | 83 | 13 | 6 | 11 | 17 | 36.1 | 56.5 | 28.8 | 10.2 | 18.6 |
| BR | 2003 | 32 | 62 | 0 | 94 | 13 | 6 | 16 | 22 | 37.2 | 40.6 | 35.5 | 9.7 | 25.8 |
| BR | 2004 | 49 | 54 | 0 | 103 | 23 | 11 | 5 | 14 | 35.9 | 46.9 | 25.9 | 20.4 | 9.3 |
| BR | 2005 | 57 | 74 | 0 | 131 | 24 | 8 | 10 | 18 | 32.1 | 42.1 | 24.3 | 10.8 | 13.5 |
|  | all years | 332 | 435 |  |  | 126 | 50 | 48 | 94 |  | 38.0 | 21.6 | 11.5 | 11.0 |
|  | all years, all ages | |  |  | 769 | 126 |  |  | 94 | 28.6 |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| HS | 1998 | 37 | 0 | 0 | 37 | 25 | 0 | 0 | 0 | 67.6 | 67.6 | NA | NA | NA |
| HS | 1999 | 38 | 12 | 0 | 50 | 28 | 7 | 1 | 7 | 70.0 | 73.7 | 58.3 | 58.3 | 8.3 |
| HS | 2000 | 29 | 51 | 0 | 80 | 26 | 45 | 27 | 51 | 96.3 | 89.7 | 100.0 | 88.2 | 52.9 |
| HS | 2001 | 38 | 12 | 0 | 50 | 23 | 12 | 0 | 12 | 70.0 | 60.5 | 100.0 | 100.0 | 0.0 |
| HS | 2002 | 41 | 9 | 0 | 50 | 29 | 6 | 1 | 6 | 70.0 | 70.7 | 66.7 | 66.7 | 11.1 |
| HS | 2003 | 53 | 87 | 0 | 140 | 33 | 26 | 7 | 28 | 43.6 | 62.3 | 32.2 | 29.9 | 8.0 |
| HS | 2004 | 64 | 6 | 0 | 70 | 38 | 2 | 0 | 2 | 57.1 | 59.4 | 33.3 | 33.3 | 0.0 |
| HS | 2005 | 41 | 12 | 0 | 53 | 17 | 8 | 4 | 10 | 50.9 | 41.5 | 83.3 | 66.7 | 33.3 |
|  | all years | 341 | 189 |  |  | 219 | 106 | 40 | 116 |  | 64.2 | 61.4 | 56.1 | 21.2 |
|  | all years, all ages |  |  |  | 530 | 219 |  |  | 116 | 63.2 |  |  |  |  |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| AL | 1998 | 20 | 0 | 0 | 20 | 15 |  |  |  | 75.0 | 75.0 | NA | NA | NA |
| AL | 2000 | 0 | 16 | 0 | 16 | 0 | 3 | 4 | 7 | 43.8 | NA | 43.8 | 18.8 | 25.0 |
|  | all years | 20 | 16 |  |  | 15 | 3 | 4 | 7 |  | 75.0 | 43.8 | 18.8 | 25.0 |
|  | all years, all ages |  |  |  | 36 | 15 |  |  | 7 | 61.1 |  |  |  |  |

# Occurance of a coho residual

During escapement sampling in October of 2010 a curious fish specimen was discovered and collected. The fish was dead on the stream bottom on the margin of a pool where coho adults aggregate in large numbers as they make their way upstream to spawning areas. The fish was coho jack size (260mm snout-fork length) but it had unusual coloration and spotting. It lacked the silvery appearance characteristic of fish returning from the marine environment and looked more trout-like, it was darker and greenish and its spotting was larger than is typical for coho and the spotting also occurred on the head and cheeks (opercles). Another irregularity was the size of the scales, they were smaller than would be expected from a jack. Though we thought it was most like a coho we investigated it further to learn more about its identity and life-history.

Samples submitted for DNA analysis identified the fish as a coho, not a hybrid of any kind. Growth patterns on the scale samples indicate that the fish never underwent growth in marine waters. The circuli numbers and spacing are consistent with freshwater growth. Also there are five annuli making this the oldest coho specimen that we have document, residual or otherwise. This came from brood year 2005 and is age 6.0, 260mm. The gonads were examined and were very underdeveloped but appeared to be testes.

The scale pattern is quite normal from the focus through the second annulus. Twelve circuli per year and nicely graduated circuli spacing indicative of seasonal changes effecting growth. After the second annulus, typically marine growth would begin, were this a common two years of freshwater rearing coho. Between the second and third annulus however, the circuli are narrower and more consistently spaced possibly indicating a reduced growth rate and less variable environmental conditions. The pattern in this area is more similar to a “lake pattern” than to a typical Berner’s river pattern. This warrants speculating that the fish found its way to a different type of habitat during this period. There is a small lake (Berner’s Lake) that attaches to the Berner’s River system; it is located about 3 Km downstream of where the carcass was recovered. Perhaps the fish was located there or perhaps some other water body where seasonal temperature variations are mitigated.

Beyond the third annulus the circuli spacing becomes more typical again with the exception that the fourth annulus is understated, it appears as a weak check. The circuli numbers continue to make sense at 10 or 11 for each year of growth before and after the fourth annulus with spacing typical of freshwater growth at Berner’s River. ­­­­

The carcass was recovered upriver of the majority of the rearing habitat and was in fact in a holding pool full of adult cohos preparing to spawn. This inspires speculation that perhaps this residual was responding instinctually to an urge to undergo a spawning process which compelled to it to swim upriver to the spawning grounds. The gonads were underdeveloped which does not help to support this scenario. However, it appears that this particular coho suffered an atypical life history that might be characterized as deviant and unfit; thus an urge towards spawning behavior while still being sexually underdeveloped may be plausible. Perhaps this urge was the cause of death indirectly by compelling the fish to depart prime rearing habitat of the lower river and head for the leaner habitat of the spawning areas. Perhaps it also suffered the loss of the urge to feed as spawners typically do, and without the fat reserves that would have been acquired by marine feeding it may have starved. The fish does not look robust in the photographs.

There is no reason to suspect a population of residual cohos inhabits the area. Several holding pools in the upper river are beach seined each fall during spawning season. The nets used select for fish larger than 25 to 30 cm depending on their circumference so residuals could escape the net if they are smaller than this threshold. However, there have been no sightings during the foot surveys or during beach seining activities. Visibility is often very good and this fish could easily be distinguished from the dolly varden that are common in the area. In the springtime the lower river is trapped using baited minnow traps (which are size selective for fish above approximately 150mm) and also using spill traps that catch fish of any size coming out of beaver ponds. There is also a small amount of sport fishing. No other occurrence of a coho residual has ever been reported at Berner’s River therefore without additional evidence, this specimen must be considered an anomaly.

|  |  |
| --- | --- |
| BR_FWresidual_flat |  |

|  |
| --- |
| Residual1A |
| Residual2A |
| Residual3A |

**AL: 1994-1998 for 2000**

Number of Observations and Percent Classified into age

From age 1 2 Total

2 13 31 44

29.55 70.45 100.00

Total 13 31 44

29.55 70.45 100.00

Error Count Estimates for age

2 Total

Rate 0.2955 0.2955

Priors 0.5000 0.5000

**BR: 1998-2004 for 2005**

Number of Observations and Percent Classified into age

From age 1 2 Total

1 188 11 199

94.47 5.53 100.00

2 4 106 110

3.64 96.36 100.00

Total 192 117 309

62.14 37.86 100.00

**HS: 1998-2004 for 2005**

Error Count Estimates for age

1 2 Total

Rate 0.0553 0.0364 0.0458

Priors 0.5000 0.5000

Number of Observations and Percent Classified into age

From age 1 2 Total

1 57 2 59

96.61 3.39 100.00

2 4 8 12

33.33 66.67 100.00

Total 61 10 71

85.92 14.08 100.00

Error Count Estimates for age

1 2 Total

Rate 0.0339 0.3333 0.1836

Priors 0.5000 0.5000

**All data: 1994-2004 for 2005**

Number of Observations and Percent Classified into age

From age 1 2 Total

1 224 34 258

86.82 13.18 100.00

2 19 103 122

15.57 84.43 100.00

Total 243 137 380

63.95 36.05 100.00

Error Count Estimates for age

1 2 Total

Rate 0.1318 0.1557 0.1438

Priors 0.5000 0.5000

|  |  |  |
| --- | --- | --- |
| **Table 1. Scale measurement and count characters calculated from intercirculus distances** | | |
| Note: variables in bold are selected for discriminant analysis. | | |
| Variable |  | Total Freshwater Annular Zone |
| **Z1** | v1 | Number of circuli in FW Annular Zone (NCFAZ) |
| **Z2** | v2 | Width of FW Annular Zone (SFAZ) |
| **Z3** | z2/z1 | Average interval between circuli (SFAZ/NCFAZ) |
| **Z4** | v3 | Number of circuli in Plus Growth Zone (NCPGZ) |
| **Z5** | v4 | Width of Plus Growth Zone (SPGZ) |
| **Z6** | z4/z3 | Average interval between circuli (SPGZ/NCPGZ) |
| **Z7** | v5 | Distance from scale focus (C0) to circulus 1 (C1) |
| **Z8** | v6 | Distance from scale focus to circulus 2 (C0 - C2) |
| **Z9** | v7 | Distance from scale focus to circulus 3 (C0 - C3) |
| **Z10** | v8 | Distance from scale focus to circulus 4 (C0 - C4) |
| **Z11** | v9 | Distance from scale focus to circulus 6 (C0 - C6) |
| Z12 | v10 | Distance from scale focus to circulus 7 (C0 - C7) |
| Z13 | v11 | Distance from scale focus to circulus 8 (C0 - C8) |
| Z14 | v12 | Distance from scale focus to circulus 9 (C0 - C9) |
| Z15 | v13 | Distance from scale focus to circulus 12 (C0 - C12) |
| Z16 | v14 | Distance from scale focus to circulus 15 (C0 - C15) |
| Z17 | v15 | Distance from scale focus to circulus 21 (C0 - C21) |
| **Z18** | v8-v6 | Distance from circulus 2 to circulus 4 (C2 - C4) |
| **Z19** | v9-v6 | Distance from circulus 2 to circulus 6 (C2 - C6) |
| Z20 | v11-v6 | Distance from circulus 2 to circulus 8 (C2 - C8) |
| **Z21** | v9-v8 | Distance from circulus 4 to circulus 6 (C4 - C6) |
| Z22 | v11-v8 | Distance from circulus 4 to circulus 8 (C4 - C8) |
| **Z23** | v9-v7 | Distance from circulus 3 to circulus 6 (C3 - C6) |
| Z24 | v12-v7 | Distance from circulus 3 to circulus 9 (C3 - C9) |
| Z25 | v13-v7 | Distance from circulus 3 to circulus 12 (C3 - C12) |
| Z26 | v14-v7 | Distance from circulus 3 to circulus 15 (C3 - C15) |
| Z27 | v12-v9 | Distance from circulus 6 to circulus 9 (C6 - C9) |
| Z28 | v13-v9 | Distance from circulus 6 to circulus 12 (C6 - C12) |
| Z29 | v14-v9 | Distance from circulus 6 to circulus 15 (C6 - C15) |
| Z30 | v14-v12 | Distance from circulus 9 to circulus 15 (C9 - C15) |
| Z31 | v15-v12 | Distance from circulus 9 to circulus 21 (C9 - C21) |
| Z32 | v14-v13 | Distance from circulus 12 to circulus 15 (C12 - C15) |
| Z33 | v15-v13 | Distance from circulus 12 to circulus 21 (C12 - C21) |
| Z34 | v15-v14 | Distance from circulus 15 to circulus 215 (C15 - C21) |
| Z35 | v16 | Distance from sixth-to-last circulus to end of zone, C(NCFAZ-6) - EOZ |
| Z36 | v17 | Distance from third-to-last circulus to end of zone, C(NCFAZ-3) - EOZ |
| **Z37** | v2-v7 | Distance from circulus 3 to end of zone (C3 - EOZ) |
| Z38 | v2-v12 | Distance from circulus 9 to end of zone (C9 - EOZ) |
| Z39 | v2-v14 | Distance from circulus 15 to end of zone (C15 - EOZ) |
| Z40 | z25/z2 | Relative width, (variable 25)/SFAZ |
| Z41 | z26/z2 | Relative width, (variable 26)/SFAZ |
| Z42 | z27/z2 | Relative width, (variable 27)/SFAZ |
| Z43 | z28/z2 | Relative width, (variable 28)/SFAZ |
| Z44 | z29/z2 | Relative width, (variable 29)/SFAZ |
| Z45 | z30/z2 | Relative width, (variable 30)/SFAZ |
| Z46 | z32/z2 | Relative width, (variable 32)/SFAZ |
| Z47 | z34/z2 | Relative width, (variable 34)/SFAZ |
| **Z48** | v18 | Number of circuli in first 1/2 of zone |
| **Z49** | v19 | Number of circuli in first 3/4 of zone |
| **Z50** | v20 | Maximum distance between two consecutive circuli |

**SAS codes**

Code-1:

/\* Convert the data format from the scale lab to the one for the discriminant analysis using SAS and collect the variables from the scale pattern\*/

%let fileid=AL\_BR\_HS; /\* Filename of input file \*/

%let filext=csv; /\* file extension \*/

libname x 'H:\SAS Splus Consulting\Juvenile coho SPA for Leon';

/\* -------------------------------------------- \*/

/\*Get import data, transpose from 1 row/fish to 1 row/cirulus\*/

**data** a;

length sampleID $ **20** tagcode $ **12** sublocation $ **16** comment $ **50**;

infile "&fileid..&filext" firstobs=**2** delimiter=',' lrecl=**1000**;

count+**1**;

input sampleID $ location $ year date $ floyno tagcode $ age length sublocation $

comment $ npairs @;

do i=**1** to npairs;/\*loop thru each data pair,output to a,1 row/circ.\*/

input zone dist @;

output;

end;

drop i count;

**run**;

**data** c; /\* change zone zone coding for age 2 so that 1fw+2fw=faz (freshwater annular zone) \*/

set a;

if age=**3** then delete;

if age=**2** then do;

if zone=**2** then zone=**1**;

if zone=**3** then zone=**2**;

end;

**run**;

**proc** **summary** data=c; /\* count up circuli and sum width by id-zone \*/

by sampleID zone;

var dist;

output out=outsum2 n=ncirc sum=totwidth;/\* output sumout=1 row/zone\*/

**run**;

**data** fazone; /\* output b = 1 row/circulus \*/

merge c outsum2(drop=\_TYPE\_ \_FREQ\_);

by sampleID zone;

**run**;

/\* Calculate basic recombinant variables: \*/

**data** cnvrt3;

set fazone;

by sampleID zone;

array v{\*} ncfaz sfaz ncpg spgz fc0\_c1 fc0\_c2 fc0\_c3 fc0\_c4 fc0\_c6

fc0\_c7 fc0\_c8 fc0\_c9 fc0\_c12 fc0\_c15 fc0\_c21

efaz\_6 efaz\_3 ncfaz1\_2 ncfaz3\_4 maxfaz;

retain v;

if first.sampleID then do i=**1** to dim(v);

v{i}=**0**;

end;

if first.zone then do;

count=**0**; sumdist=**0**;

end;

count+**1**;

sumdist+dist;

if zone=**1** then do;

if count>**1** and dist>maxfaz then maxfaz=dist;

if count=**1** then fc0\_c1=sumdist;

else if count=**2** then fc0\_c2=sumdist;

else if count=**3** then fc0\_c3=sumdist;

else if count=**4** then fc0\_c4=sumdist;

else if count=**6** then fc0\_c6=sumdist;

else if count=**7** then fc0\_c7=sumdist;

else if count=**8** then fc0\_c8=sumdist;

else if count=**9** then fc0\_c9=sumdist;

else if count=**12** then fc0\_c12=sumdist;

else if count=**15** then fc0\_c15=sumdist;

else if count=**21** then fc0\_c21=sumdist;

if ncirc-count=**6** then efaz\_6=totwidth-sumdist;

else if ncirc-count=**3** then efaz\_3=totwidth-sumdist;

if sumdist le **.5**\*totwidth then ncfaz1\_2=count;

else if sumdist le **.75**\*totwidth then ncfaz3\_4=count;

if last.zone then do;

ncfaz=ncirc; sfaz=totwidth;

end;

end;

else if zone=**2** then do;

ncpg=ncirc; spgz=totwidth;

end;

if last.sampleID then output;

**run**;

**data** x.&fileid.\_FAZ;

set cnvrt3;

array v{\*} ncfaz sfaz ncpg spgz fc0\_c1 fc0\_c2 fc0\_c3 fc0\_c4 fc0\_c6

fc0\_c7 fc0\_c8 fc0\_c9 fc0\_c12 fc0\_c15 fc0\_c21

efaz\_6 efaz\_3 ncfaz1\_2 ncfaz3\_4 maxfaz;

z1=v{**1**}; z2=v{**2**}; z3=z2/z1; z4=v{**3**};

z5=v{**4**}; z6=z4/z3; z7=v{**5**}; z8=v{**6**};

z9=v{**7**}; z10=v{**8**}; z11=v{**9**}; z12=v{**10**};

z13=v{**11**}; z14=v{**12**}; z15=v{**13**}; z16=v{**14**};

z17=v{**15**}; z18=v{**8**}-v{**6**}; z19=v{**9**}-v{**6**}; z20=v{**11**}-v{**6**};

z21=v{**9**}-v{**8**}; z22=v{**11**}-v{**8**}; z23=v{**9**}-v{**7**}; z24=v{**12**}-v{**7**};

z25=v{**13**}-v{**7**}; z26=v{**14**}-v{**7**}; z27=v{**12**}-v{**9**}; z28=v{**13**}-v{**9**};

z29=v{**14**}-v{**9**}; z30=v{**14**}-v{**12**};z31=v{**15**}-v{**12**}; z32=v{**14**}-v{**13**};

z33=v{**15**}-v{**13**};z34=v{**15**}-v{**14**}; z35=v{**16**}; z36=v{**17**};

z37=v{**2**}-v{**7**}; z38=v{**2**}-v{**12**}; z39=v{**2**}-v{**14**}; z40=z25/z2;

z41=z26/z2; z42=z27/z2; z43=z28/z2; z44=z29/z2;

z45=z30/z2; z46=z32/z2; z47=z34/z2; z48=z18;

z49=z19; z50=z20;

keep sampleID location year date floyno tagcode age length sublocation comment z1-z50;

**run**;

Code-2

%let loc = BR; /\*location of fry; loc can be AL, BR, or HS\*/

**data** d1; /\*get rid of useless variables.\*/

set x.Al\_br\_hs\_faz (drop = tagcode sublocation comment date floyno length z12-z17 z20 z22 z24-z36 z38-z47);

**data** x.&loc; /\*analyze Hugh Smith Lake data\*/

set d1;

if location = "&loc";

**data** x.knowns; /\*generate subdata set, leave year 2005 data for test\*/

set x.&loc;

if year = **2005** then delete;

**data** x.unknowns; /\*2005 data to be classfied\*/

set x.&loc;

if year = **2005**;

**run**;

Code-3

/\*stepwise discriminant to select variables that are used for discriminant analysis \*/

**proc** **stepdisc** data=x.knowns;

class age;

var z1-z11 z18 z19 z21 z23 z37 z48-z50;

**run**;

Code-4

/\*use this calibration information to classify observations in testdata\*/

**proc** **discrim** data=x.knowns testdata=x.unknowns testlist;

class age;

var z3 z6 z8 z21 z23 z37 z50;

title 'linear discriminant analysis';

**run**;

# results

# discussion

# recommendations

# references cited

# Appendix