Back-Calculated Fish Lengths, Percentages of Scale Growth, and Scale Measurements for Two Scale Measurement Methods used in Studies of Salmon Growth

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Abstract: Scale measurements, percentages of annual scale growth, and back-calculated fish lengths were examined for the INPFC method (75° anterior reference line) and Traditional method (anterior-posterior reference line) on scales of chum salmon (*Oncorhynchus keta)* for three ages, two stocks, and two brood years. Mean measurements of annual increments of scale growth were greater for the INPFC method than the Traditional method. Differences in percentages of scale growth and back-calculated fish lengths for the two methods were tested using the Hotelling one-sample T^2 test. Percentages of annual scale growth differed significantly ($p \leq 0.003$) between the two methods for seven of nine tests. Mean percentage of annual scale growth was greater in the first year for the Traditional method and in the final year for the INPFC method; intermediate years were similar. Back-calculated fish lengths computed by the Fraser-Lee method differed significantly between the INPFC and Traditional method ($p \leq 0.004$) for seven of nine tests. Although differences in back-calculated lengths of the two methods were statistically significant, the biological differences were small (~1 cm).

INTRODUCTION

Scales from many species of fish are commonly used to estimate age, growth history, spawning history, and racial differences. Also, when the scale radius is proportional to the length of the fish, size at younger ages can be estimated by back-calculation using scale measurements (Van Oosten 1928; Hile 1936). Back-calculations can be used to trace the effects of winter oxygen levels (Casselman and Harvey 1975), fishing pressures (Nicholls 1958), and food consumption (Weatherly 1959) on growth rate.

The number of circuli in the first ocean zone of Pacific salmon (*Oncorhynchus* spp.) scales is often used to identify racial differences. The circuli may be counted along several different orientations on the scale; in back-calculation studies the most commonly used line of measurement is the anterior-posterior line through the focus of the scale (Fig. 1), which we refer to as the Traditional method.

LaLanne (1963) observed in chum salmon (*O. keta*) that the Traditional method circuli were often broken or discontinuous, so he developed an alternative line of count. This line was developed for racial studies on chum salmon for the International North Pacific Fisheries Commission (INPFC). LaLanne (1963) determined that a line that bisects the focus at 75° from a line drawn between the focus and shortest distance to where the first annulus enters the translucent exposed portion of the scale provided circuli counts with the least variation (Fig. 1); we call this the INPFC method. Both the Traditional and INPFC methods are used in scale growth studies of chum salmon.

To understand the differences between the Traditional and INPFC methods, we compared scale measurements, percentages of annual scale growth, and back-calculated fish lengths. This study presents results for different stocks, brood years, and ages.

MATERIALS AND METHODS

Length measurements and scales were collected by author Helle from chum salmon in August 1985 and 1992 at Fish Creek, located at the head of Portland Canal in southern southeastern Alaska (U.S.A.) and in December 1985 at Quilcene National Fish Hatchery, in Hood Canal, Washington (U.S.A.). Scales were taken from the "preferred area" of the body (Anas 1963) and placed on gummed cards with the reticulated side facing up. Lengths were taken from the middle of the eye to the end of the hypural plate (MEHP).

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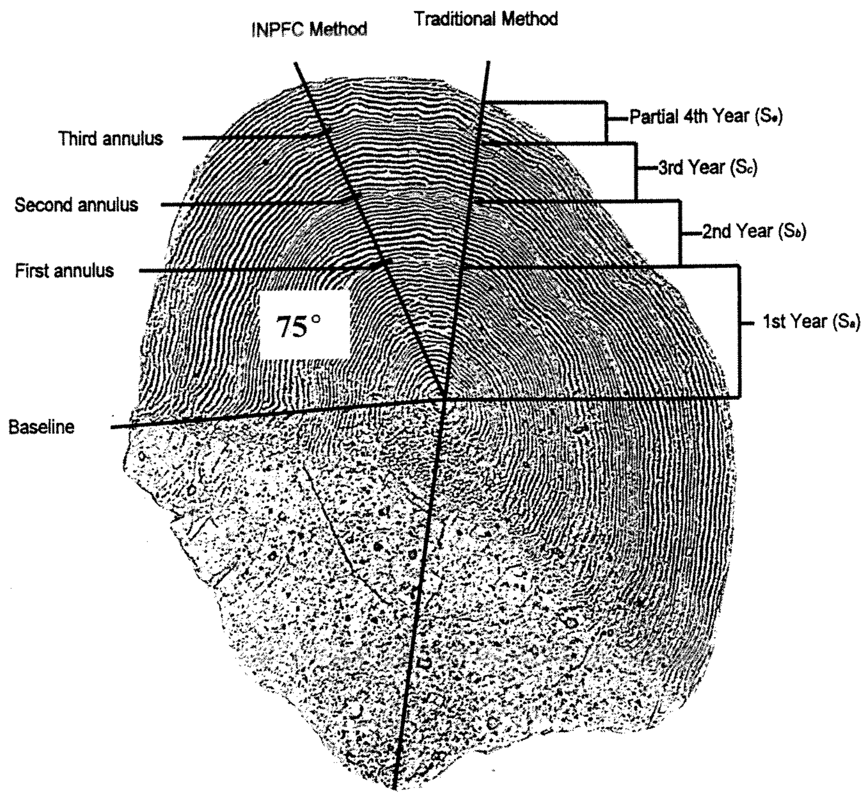
Fig. 1. Scale of an ocean-age 3 male chum salmon illustrating the INPFC method (75° reference line) and Traditional method (anterior-posterior reference line), with years of growth.



In the laboratory, scales were pressed onto plastic acetate cards using a heated hydraulic press to produce impressions of the scales (Arnold 1951) and viewed on an Eberbach[1] projector at a magnification of 80x. Ages were designated using the European method, but excluding the number of freshwater years and the decimal point (Koo 1962); therefore we merely present number of winters spent at sea (ocean age). Ocean age was read by counting the number of true annuli on the scale. We examined only scales of ocean-age 2, 3, and 4 male chum salmon. Scales were measured from the focus to the first annulus (S_a), first to the second annulus (S_b), second to third annulus (S_c), third to the fourth annulus (S_d), and from the last annulus to the edge (S_e) using the INPFC and Traditional methods. Measurements were then converted to actual size by dividing by 80 (the magnification).

Back-calculation Method

Back-calculations were based upon a linear regression model developed by Fraser (1916) and Lee (1920), which assumes that fish length is directly proportional to scale radius (Dahl 1909). The model can be written as

$$L_i = c + (L_T - c)*(S_i / S_T),$$

where i = age at the time of annulus formation,

c = length of fish at the onset of scale formation,

L_T = fish length at capture,

L_i = fish length at time of annulus formation,

S_T = scale radius at capture, and

S_i = scale radius at time of annulus formation (ocean age).

Scale radii at time of annulus formation (S_i) were as follows: first annulus or ocean age-1 ($S_1 = S_a$), second annulus or ocean age-2 ($S_2 = S_a+S_b$), third annulus or ocean age-3 ($S_3 = S_a+S_b+S_c$), and fourth annulus or ocean age 4 ($S_4 = S_a+S_b+S_c+S_d$).

In estimating length of fish at time of scale formation (c), we used length measurements taken from fry before they developed scale circuli. Helle (1979) reported that chum salmon fry (n = 29) in the intertidal zone at Olsen Creek, Prince William Sound, had scales but no circuli. On average, these fish were 46 mm from the tip of the snout to the fork of the tail (TSFT). In the lab we measured TSFT and MEHP of chum salmon fry (n = 50) before they were released from Douglas Island Pink and Chum Fish Hatchery,

[1]Reference to trade names does not imply endorsement by the National Marine Fisheries Service, NOAA.

Juneau, Alaska in 1999 and developed a regression model: MEHP = 0.85*TSFT-2.9; $r = 0.99$. Using the regression model, TSFT (46 mm) was converted to MEHP (36 mm) and used as common intercept (c) for the INPFC and Traditional methods in the back-calculation model.

Graphical and Statistical Method

Measurements of annual scale growth (S_a, S_b, S_c, S_d, S_e), percentage scale growth within years (S_a/S_T *100, S_b/S_T*100, S_c/S_T*100, S_d/S_T*100, S_e/S_T*100), and back-calculated lengths at each ocean age (L_1, L_2, L_3, and L_4) were evaluated using vertical error bar charts showing mean and 95% confidence intervals.

The back-calculated fish lengths for the INPFC method and Traditional method can be considered as two data sets from which we draw inferences. Each data set consists of vectors of several lengths per fish (for example $\boldsymbol{L} = (L_1, L_2, L_3)'$ for ocean-age 3 fish), where the subscripts represent ocean ages. The data sets themselves are not independent from each other, but are repeated-measures data; that is, back-calculated fish lengths for the two axes are computed from the same fish. Because the lengths calculated at each ocean age (2–4) are probably dependent on each other, the data lend themselves to multivariate analysis. To determine whether the back-calculated lengths computed from the two scale methods differed significantly from each other, we compared the paired vectors of lengths ($\mathbf{L_I}$ and $\mathbf{L_T}$) for all the fish, where I = INPFC method and T = Traditional method. Differences were computed between paired back-calculated lengths, $\mathbf{d} = \mathbf{L_I} - \mathbf{L_T}$. If the $\mathbf{d}$'s are considered to be sampled from a multivariate normal population with mean δ, then $\bar{\mathbf{d}}$ can be compared to δ. Specifically, we computed a multivariate likelihood ratio statistic (Mardia et al. 1979) to test the null hypothesis: H_0: $\delta = \delta_0$, $\boldsymbol{\Sigma}$ unknown, where $\delta_0 = \mathbf{0}$. This test is also known as a Hotelling one-sample T^2 test. Assuming the data are multivariate normally distributed, test statistics can be compared to an F distribution to determine significance probability. We used a battery of normality tests (Aitchison 1986) on the data to verify the assumption of multivariate normality.

The sets of percentages of annual scale growth for the two scale methods were analyzed similarly. Each data set consisted of vectors of three percentages (for example $\boldsymbol{S} = (S_a/S_T *100, S_b/S_T*100, S_c/S_T *100)'$ for ocean-age 3 fish). To determine whether the percentages of scale growth computed from the two scale methods differed significantly, a Hotelling one-sample T^2 test similar to the one described above was performed.

RESULTS

Mean back-calculated fish lengths for all ages, stocks, and brood years were greater for the Traditional method than for the INPFC method (Fig. 2). Mean calculated lengths for the Traditional method were 229–249 mm at ocean-age 1, 395–405 mm at ocean-age 2, and 501–504 mm at ocean-age 3. Mean calculated lengths from the INPFC method were 217–240 mm at ocean-age 1, 390–402 mm at ocean-age 2, and 495–500 mm at ocean-age 3. Differences in mean back-calculated fish lengths between scale methods were 3–9 mm at ocean-age 1, 1–9 mm at ocean-age 2, 2–10 mm at ocean-age 3, and 0–15 mm at ocean-age 4. Differences in back-calculated length between the scale methods varied more between stocks than between brood year or age of the same stock (Fig. 2).

After performing a battery of normality tests (Aitchison 1986) on the back-calculated lengths, we concluded the data were multivariate normally distributed. We then tested whether the lengths back-calculated from increments using the two scale methods differed significantly, using a Hotelling one-sample T^2 test. Lengths calculated from the two methods differed significantly ($p \leq 0.004$) for all stocks, brood years, and ages except brood-year 1981 ocean-age 4 fish from Fish Creek ($p = 0.27$) and Quilcene National Fish Hatchery ($p = 0.33$).

Mean percentages of annual scale growth were greater for the Traditonal method in the first ocean year (~2%) and greater for the INPFC method in the last ocean year (~2%) (Fig. 3). For intermediate years, difference in percentages of annual scale growth varied little between methods. Normality tests were performed similarly on the scale percentages. Because the percentages did not deviate greatly from the multivariate normal distribution, we assumed the data were multivariate normally distributed. Differences in percentages along the two axes were then tested using a Hotelling one-sample T^2 test. Percentages for the two methods differed significantly ($p \leq 0.003$) for all stocks, brood years, and ages except for brood-year 1981 ocean-age 4 fish from Quilcene National Fish Hatchery ($p = 0.30$) and Fish Creek ($p = 0.28$). Note that sample size from Quilcene National Fish Hatchery was small ($n = 13$).

Mean measurements of annual increments of scale growth were greater for the INPFC method: 0.01–0.11 mm in the first year (1–9%), 0.05–0.13 mm in the second year (6–13%), 0.00–0.06 mm in the third year (0–14%), 0.20–0.50 mm in the fourth year (10–14%), and 0.04–0.15 mm in the last year (7–24%). Scales of Quilcene fish had greater differences between scale methods than scales of Fish Creek fish (Fig. 4).

Fig. 2. Fish length at each ocean age, or annulus, calculated from chum salmon scale measurements using the INPFC and Traditional methods. Circles equal means and bars are 95% confidence intervals of the mean. Sample size is in parentheses.

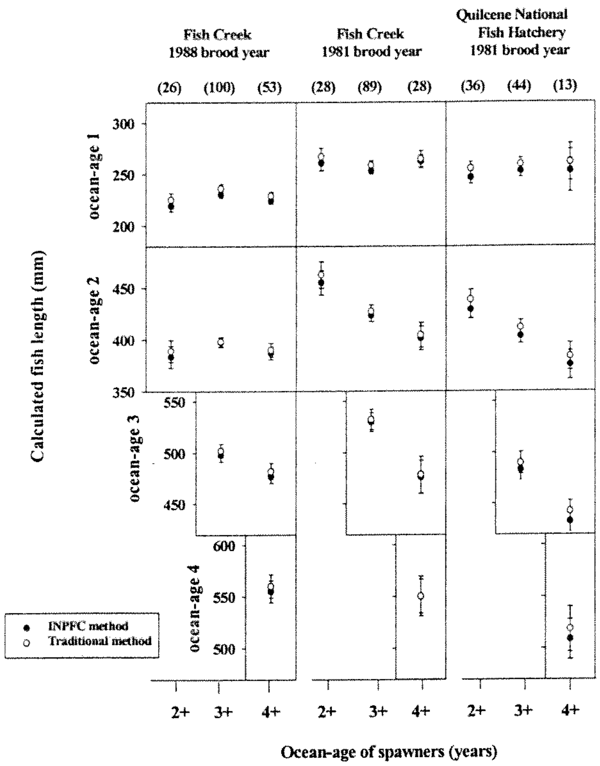


Fig. 3. Percentage of scale growth during each marine year for the INPFC method and Traditional method on scales of chum salmon. Circles are means and bars are 95% confidence interval of the mean. Sample size is in parentheses.

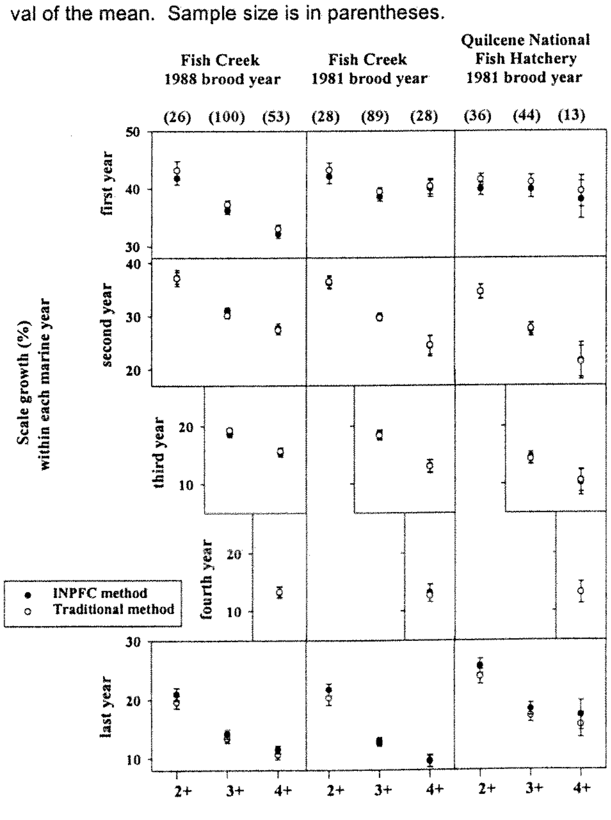
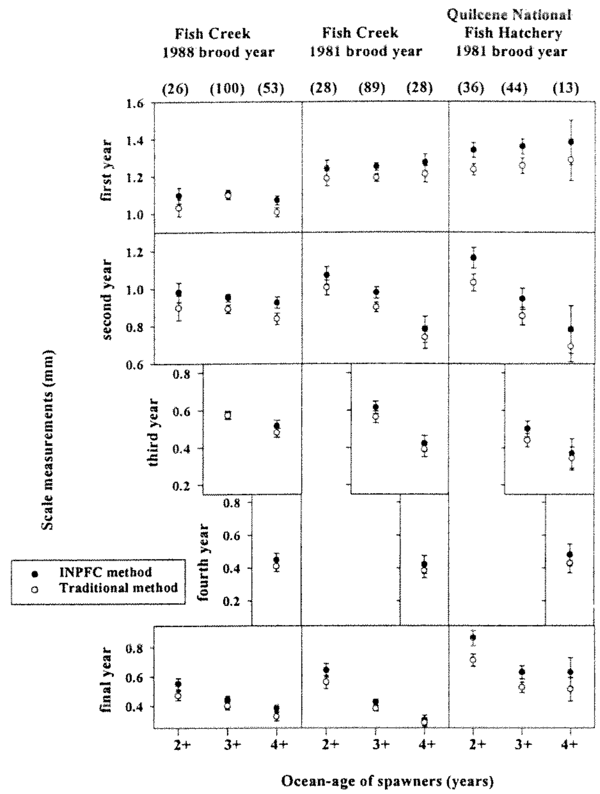


Fig. 4. Measurements of annual increments of growth for INPFC and Traditional methods on scales of chum salmon. Circles are means and bars are 95% confidence intervals of the mean. Sample size is in parentheses.



DISCUSSION

The choice of scale method (position of the reference line on the scale) influenced the back-calculated fish lengths (BCFLs), percentages of annual scale growth, and scale measurements. Differences in the two methods were greater between stocks than between brood years or ages.

In other species, scale method also influenced scale measurements, circuli counts, and back-calculated fish lengths. For Lake Herring (*Leucichthys artedi)*, Van Oosten (1928) found significant differences between BCFLs for the lateral-radial axis and antero-lateral axis (Traditional method), both between and within races. On scales of sockeye salmon smolts, scale radius and circuli counts differed between the longest axis, dorsal 20° radial axis, and ventral 20° radial axis (Clutter and Whitesel 1956). In northern pike (*Esox lucius*), scale radius varied between the anterior axis, lateral axis, and posterior axis (Casselman 1990).

Calculated lengths corresponded well with empirical lengths of immature chum salmon at sea (Carlson et al. 1998), except at ocean-age 1. Calculated lengths at ocean-age 1 were 22–44% shorter than the empirical lengths for both methods. Are larger fish being selected against after the first year at sea, or are we making an invalid comparison? We question the validity of comparing BCFLs of single-stock fish to measured lengths of mixed-stock fish from the high seas because of variation in growth between stocks (Hile 1970; Helle 1984). Back-calculations could be validated using mark-recapture techniques on the high seas.

In studies validating Fraser-Lee BCFLs, lengths at earlier ages were commonly underestimated. For bluegill (*Lepomis macrochirus*), and bluegill X sunfish (*L. cyanellus*) hybrid, the low estimates of lengths at earlier ages were attributed to difficulties in recognizing the second annulus (Klumb and Bozek 1999). We were able to recognize the location of the annuli in our study. For rainbow trout (*O. mykiss*), the underestimates were attributed to the influence of the intercept as fish length increased (Davies and Sloane 1986). If we had used a larger intercept (*c*), the calculated lengths at ocean-age 1 would have been larger for INPFC and Traditional methods, but the difference between the methods would have decreased by only a few millimeters.

The Traditional method usually provided greater estimates of BCFLs at each ocean age (Fig. 2) than the INPFC method. In most cases, the Traditional method had larger additive percentages of scale growth at each consecutive annulus than the INPFC method. Because Fraser-Lee BCFLs are based on the addition of consecutive percentages of annual scale growth, the BCFLs were larger for the Traditional method at each ocean age.

CONCLUSION

The selection of a particular scale method (orientation of line of measurement) for analyzing growth of chum salmon may significantly affect BCFLs, measured percentage of scale growth, and scale measurements. Although differences in mean BCFLs between the methods were statistically significant, the biological differences were small (~1 cm). These results support the need for standardization of a scale measurement method.

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