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Seattle, Washington 98195

CHUM SALMON OCEAN RANCHING STUDIES AT THE BIG BEEF CREEK FISH RESEARCH STATION

FINAL REPORT

Project Period: November 1, 1974 to September 30, 1975

Prepared by: Bruce P. Snyder
and
Steven L. Schroder

Ernest O. Salo
Principal Investigator

Cooperator: George W. Farwell
Vice President
for Research

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Approved: Robert L. Burgner
Robert L. Burgner
Director, FRI

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CHUM SALMON OCEAN RANCHING STUDIES AT THE BIG BEEF CREEK FISH RESEARCH STATION

(Final Report for the Period November 1, 1974 to September 30, 1975)

GENERAL INTRODUCTION

Objectives

This report examines several problems related to chum salmon (*Oncorhynchus keta*) enhancement programs which must rely on artificial propagation. The principal areas of discussion include: 1) an examination of mate selection patterns in chum salmon--a basis for decisions related to the development of breeding programs for this species, 2) maternal and paternal influences on the development and growth of chum salmon larvae and fry, 3) effects of various types of substrates on the efficiency of yolk material utilization in chum salmon larvae incubated within Heath Tecna incubators, 4) development of a prototype incubation box for chum salmon eggs and larvae, and 5) the trials and tribulations of chum salmon culture in the hatchery and in rearing pools.

Observations related to these questions were made either at the University of Washington's Big Beef Creek Fish Research Station (Fig. 1) or at a neaby artesian water source approximately 2 kilometers from the station.

Experimental Fish

The chum salmon used for the investigations originated from Big Beef Creek which flows into the east shore of Hood Canal (Kitsap County, Washington state). Mature chum salmon enter the stream from early September through mid-January, with peaks of abundance occurring in early October (early run), mid-November (middle run), and late December (late run).

As the fish migrate into Big Beef Creek, they are captured in one of two traps and processed as described by Schroder (1973) and Koski (1975). Each fish used in the present studies was examined for maturity, anesthetized in MS-222 (tricane methanesulfonate), weighed, measured, and aged.

Fish which were used as broodstock for the hatchery investigations were often spawned the same day of their capture. Immature fish were held in pens until ripe (usually less than 4 days) and spawned at that time.

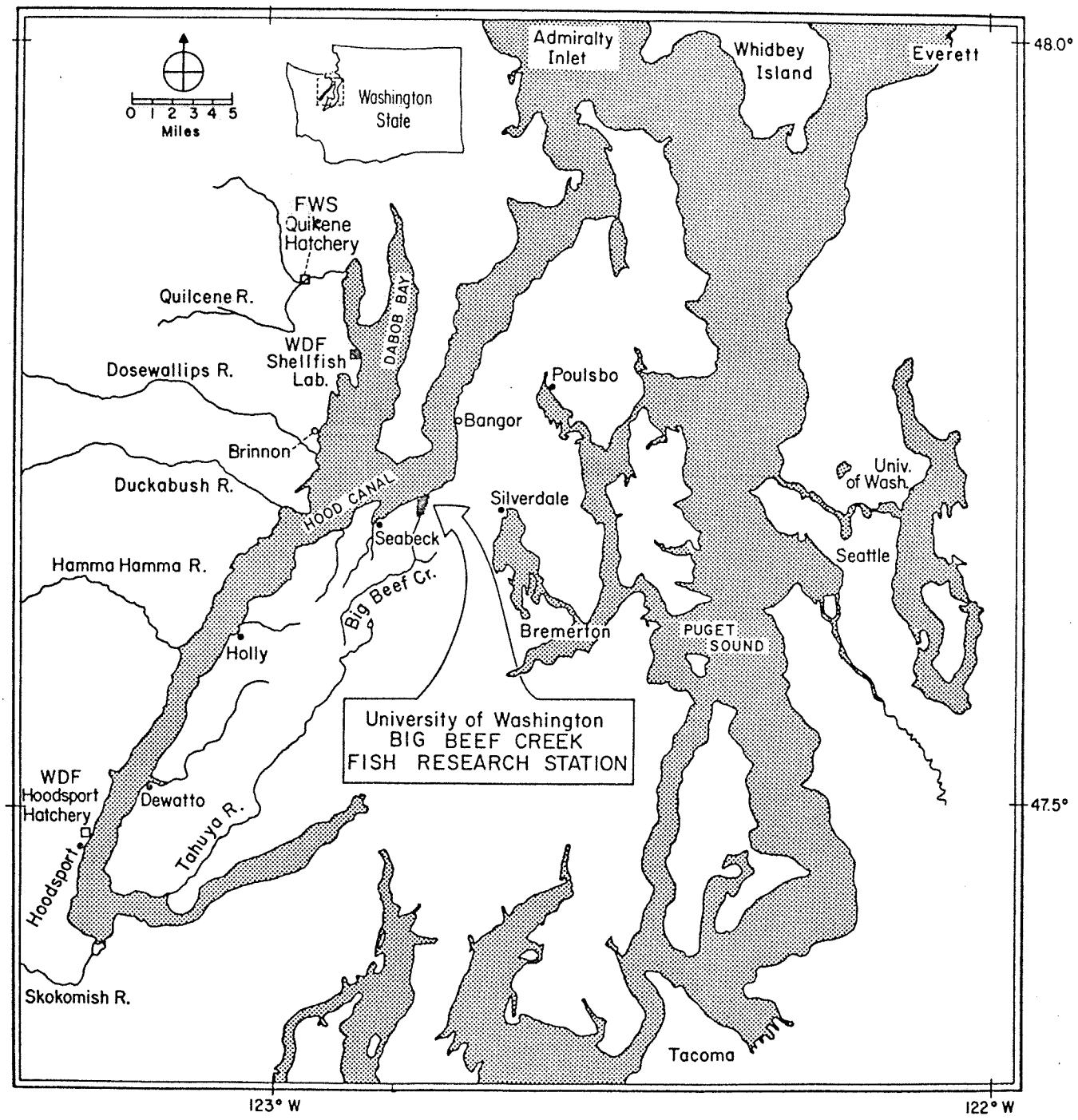


Fig. 1. Location of the University of Washington's Big Beef Creek Fish Research Station.

I. AN EXAMINATION OF MATE SELECTION PATTERNS IN CHUM SALMON--A BASIS FOR DECISIONS RELATED TO THE DEVELOPMENT OF BREEDING SCHEMES FOR THIS SPECIES

Introduction

Two basically different systems of fish culture are possible; these are confined (intensive culture) and unconfined (extensive culture) systems (Helle, 1976). The genetic strategies needed for each system are different. Breeding programs similar to those developed in agriculture (e.g., development of inbred lines, heterosis, selective breeding, etc.) would be applicable if the fish were to be confined in ponds, raceways, etc., for their entire lifetimes (intensive culture). If, on the other hand, the animals are to be released into their natural habitat zones, then the above techniques, which tend to reduce genetic variability, may be more detrimental than beneficial to the enhanced stock (Helle, 1976). For clearly, animals possessing a high degree of adaptive genetic variability will have a distinct advantage over those which do not, when both types are exposed to the fluctuating natural environment (Mayr, 1970; and Helle, 1976).

Consequently, the first thing of interest to a prospective or practicing fish culturist intending to use an unconfined system of aquaculture (such as an "ocean ranch") should be the comprehension of how and, more importantly, why genes are exchanged the way they are within demes of the species he is attempting to propagate. Yet, this concern is often ignored, and frequently at production facilities, assortative patterns of mate selection are employed that have no proven relationship with biological reality, but instead are based upon the intuitive knowledge and "art" of the fish culturist. One consequence of such poorly conceived breeding programs is a loss in genetic diversity in the cultured stock, and the subsequent creation of genetically uniform strains of fish. Such strains have been identified in hatcheries (Calaprice, 1969; Simon, 1972; and Helle, 1976) and yet, little attention has been paid to the genetic implications of these findings (Helle, 1976).

We felt that an understanding of how genes are exchanged in naturally reproducing populations of chum salmon would provide valuable information in the development of breeding schemes for ocean ranches utilizing this species. Our attempts to examine this phenomenon were restricted to observations made on mature salmon that were placed into sections of the Big Beef Creek controlled-flow spawning channel (for a detailed account of these experiments, *see* Schroder, 1975).

Methods and Materials

Spawning Channel

Eight 3.05-m x 15.2-m sections of the spawning channel were used to examine mate selection patterns under various spawner densities and sex ratio regimes. Each section of the channel had a 0.25% gradient (.25 m/100 m), was filled with 75 cm of coarse (.8-cm to 6.25-cm) stream gravel, and was equipped with a 3-m high observation wall supplied with viewing ports. Water velocity was kept at 22.5 cm/sec and depth at 30 cm throughout the experiments. Further details concerning the construction, design, and additional experimental uses of the channel can be found in Beall (1972), Schroder (1973), and Koski (1975).

Observations of Fish in the Spawning Channel

Observations were made only during daylight hours and were primarily concerned with determining the reproductive status of both males and females in each experimental section of the channel. Since each fish had an identifying set of tags, it was possible to determine the size ratio and ages for each mating pair observed.

Results and Discussion

Basic Behavioral Patterns of Spawning Chum Salmon

In attempting to discover the patterns of mate selection that may exist in chum salmon, it was necessary to examine the social behavior of adults while they interacted on the spawning grounds. Fortunately, it appears that enough of the behavioral repertoire of this species is visually perceivable so that patterns can be recognized by simple observations.

We found that the basic behavioral patterns of each sex were distinctly different. Females are territorial and may utilize visual, tactile, and chemical clues in finding a suitable spawning site. Once a female has established a territory, she will construct a series of three to six discrete nests and remain in close proximity to them until she dies. In general, females are strongly substrate oriented, with the majority of their attention being directed to nest or mound (the collective burial of all the nests under one large mound of gravel) construction. Prolonged aggressive interactions among females are rare, with the preponderance of intrasex aggression occurring among females less than 2 m away (Schroder, 1973).

Males do not establish permanent territories but tend to be mobile, moving from one territorial female to another. Aggression among males is common, and prolonged ritualistic challenges and battles may occur among similarly sized rivals, usually directly over the contested female's nest site, often preventing other males from courting and spawning with her. After spawning, a male will remain by a female for approximately 20 minutes or less (often drifting away and returning at infrequent intervals) before he abandons her, possible locating another whose nest is nearer completion and hence, closer to spawning.

Both sexes tend to be opportunistic and promiscuous and thus, it is possible for a female's eggs to be fertilized by four or more males. If there are more males than females on the spawning grounds, or if the population of spawners is numerous enough to limit the numbers of females that can find and hold territories (producing a situation where males will outnumber females with territories), then more than one male quite often spawns with a female (Fig. 2). Consequently, so-called satellite males can increase the number of sexual partners that may fertilize portions of a female's egg complement.

Dominant, or alpha, males can easily be distinguished from satellite males by their color patterns, courting movements, and close downstream proximity to a female. When observations were made to determine whether chum salmon paired in predictable patterns, only the size of the alpha male (often only one male was present) was used to calculate the size ratio that existed between a courting pair. Furthermore, it sometimes happens that a male may fertilize more than one clutch of eggs a female deposits. When such cases were observed, the size ratio of the pair was used as many times as it occurred.

The results of these observations are Shown in Figs. 3a to 3d. Each figure indicates what the expected (random) and observed frequencies of pairings were among fish of various size ratios (male body weight:female body weight). To test whether these frequencies were statistically different, a Goodness-of-Fit Test (G statistic, Sokal and Rohlf, 1969) was used. This test indicated that when the sex ratio was at parity or when there were more males than females, pairing did not occur randomly. Visual inspection of the figures suggests that chum salmon preferentially pair with individuals of approximately the same size. In most cases, if a male was smaller than a female, he was rarely able to exclusively court or spawn with her. Conversely, males that were larger than their prospective mates were often successful. By comparing the size ratios that existed between males and females originating from various brood years, to those which existed between the observed pairs, it was found that mate selection may also be assortative with respect to age as well as size.

Fig. 3c illustrates that subtle changes may also occur in the size ratios of pairs that form when spawner density is moderate ($\leq 4m^2$/female) and there are twice as many males as females. Under these circumstances,

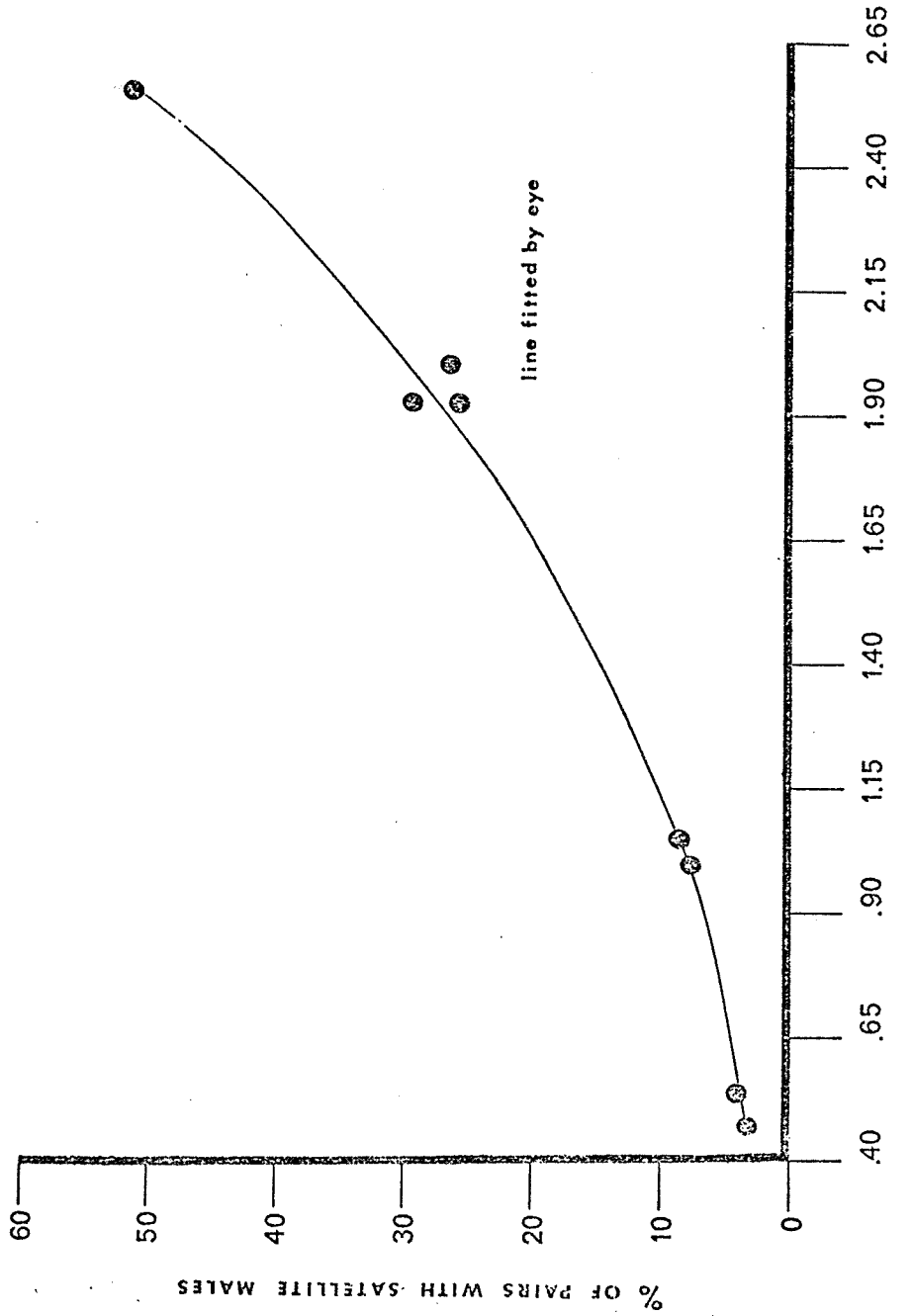


Fig. 2. The occurrence of satellite males (socially subdominant males) behind courting pairs of chum salmon at various sex ratios.

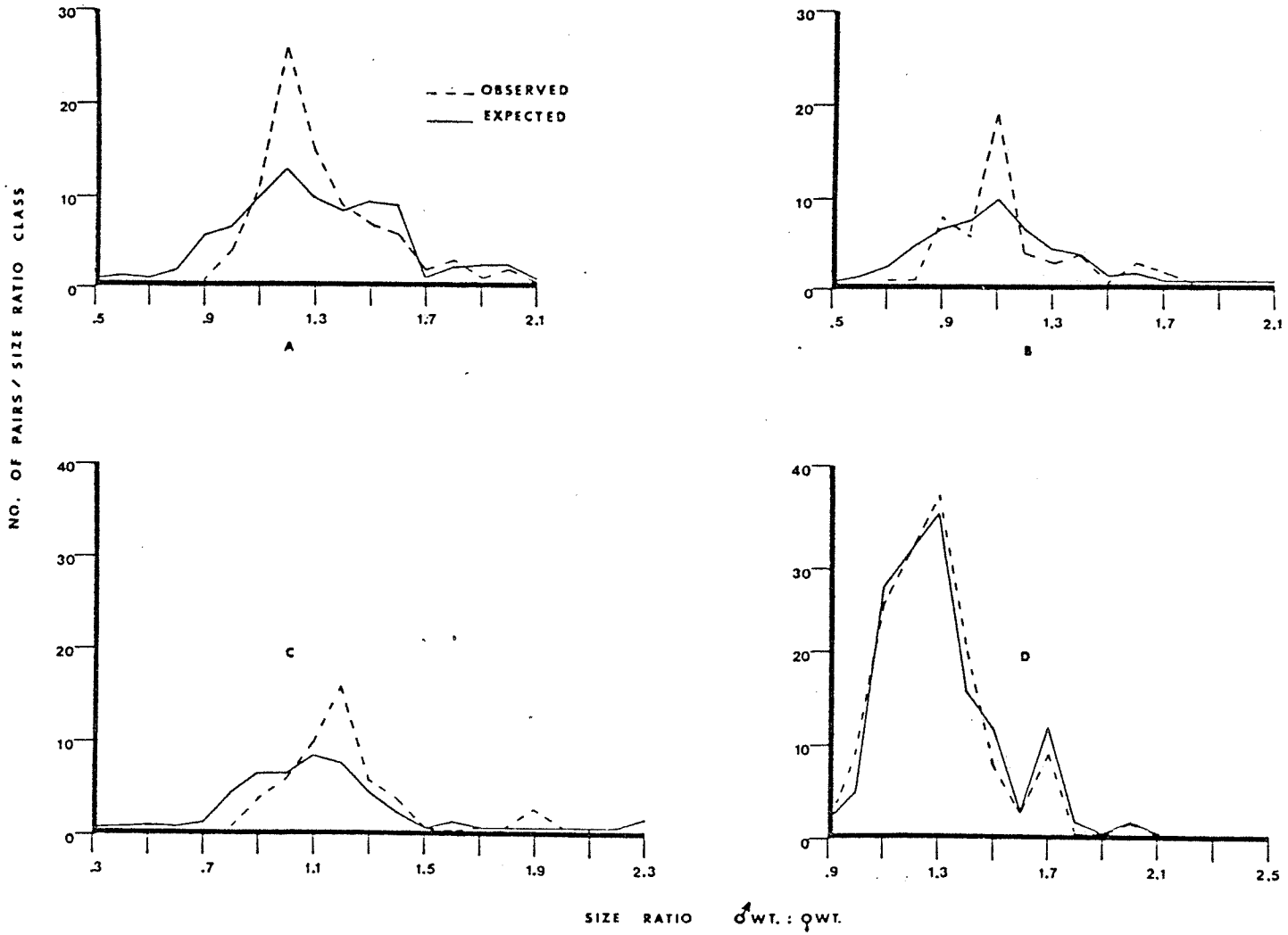


Fig. 3. The expected (random) and observed frequencies of pairings between mating chum salmon of various size ratios (♂ body weight:♀ body weight) under a variety of sex ratios and spawner densities. The sex ratio in 3a and 3b was 1:1 with a spawner density of 1.55 $m^2/♀$. In 3c, the sex ratio was 2♂♂:1♀ and the spawner density equaled 3.58 $m^2/♀$, while in 3d the sex ratio was 1♂:2♀♀ and each female was allotted 1.5 m^2 of spawning substrate.

competition among male rivals is intense and consequently, the relative size and strength of a male are given additional importance. However, when spawner density is low ($\geq 4m^2$/female) and the number of gravid females holding territories exceeds the number of males present, then mate selection tends to lose its assortative properties and mating becomes random (Fig. 3d).

The adaptive significance of mate selection patterns can only be speculated upon. Two major opposing selection pressures which deal with the degree of genetic diversity within populations appear to be in dynamic conflict. Briefly, these pressures can be identified as 1) those which cause a high degree of genetic diversity (giving succeeding generations a greater genetic background to interact with a fluctuating environment) and 2) those which are designed to reduce the wasteful production of locally inferior genotypes. An equilibrium of these forces is maintained in populations because extreme genetic variability is just as detrimental as genetic uniformity (Mayr, 1970).

Mating in chum salmon (although variable) rarely occurs in a random fashion. It seems reasonable to ask whether certain types of matings will provide a greater reproductive value to the spawners than other possible gene combinations that might have been selected. It may be possible that the behavioral activities we have observed are designed to perpetuate discrete life-history strategies within co-existing subpopulations of fish. Our analyses of this possibility are incomplete so, until the evolutionary implications of the patterns we observed are understood, we recommend that the genetic variability of ocean ranch stocks be maintained by using gametes randomly chosen from individuals representing the entire stock (Helle, 1976).

II. MATERNAL AND PATERNAL INFLUENCES ON THE DEVELOPMENT AND GROWTH OF CHUM SALMON LARVAE AND FRY

Introduction

In the previous section, we suggested that mate selection patterns may ultimately be caused by the differential survival of offspring produced by particular types of matings (gene exchanges). It would be of great practical value to determine whether maternal or paternal characteristics influence the fitness of progeny in a predictable and consequently exploitable fashion. The parental characteristics which we examined included egg sizes, and male and female age.

Egg Characteristics of 3- and 4-year-old chum salmon

Two hundred twenty-one females (forty-seven 3-year-olds and one hundred seventy-four 4-year-olds) were used to examine the characteristics of eggs produced from 3- and 4-year-old fish originating from the middle run of Big Beef Creek. The entire egg complement of each female was removed and weighed to the nearest g on a Chatillon 6-k x 10-g balance. After the eggs were weighed, a sample of 60 to 200 eggs was withdrawn and weighed on a Mettler P 1200 balance to the nearest 0.01 g. These two weights were used in the following algebraic formula to estimate the fecundity of each female:

$$\text{fecundity} = \frac{(\text{no. of eggs/sample})\ (\text{total egg wt})}{\text{weight of the sample}}$$

The eggs from each sample were then allowed to water harden for 12 hours before subsamples were measured and weighed. Three subsamples of 20 eggs from each sample were measured to the nearest .5 mm on a board held by a stand at a 25-degree angle. The subsamples of measured eggs were blotted dry and weighed to the nearest .01 g on a Mettler P 120 balance, placed in a Precision Thelco Model 17 drying oven, and baked for 24 hours at 97°C. After drying, the eggs were placed in a vacuum dessicator with anhydrous calcium chloride and allowed to cool before they were reweighed on the Mettler P 120 balance.

Fish Culture

To examine the possible effects of parental age and size on the developmental rates and growth patterns in alevins and fry, two sets of controlled breeding experiments were performed. In both experiments, the female parents were anesthetized in MS-222, had their caudal peduncles severed, and were bled thoroughly before any eggs were removed. Milt from selected males was added to the eggs and the spawn was then transported approximately 2 km from the research station to an artesian water source. The eggs were fertilized at this location just prior to their placement into standard Heath incubator trays. After the eggs had reached the "eyed stage" of development, they were "shocked" and mortalities from each cross were removed. The remaining live eggs were returned to Heath trays which had been modified by the attachment of an Astroturf substrate to the bottom screen. A flow of 11 liters/min was maintained in each Heath tray and the water temperature ranged from 8.7 C to 8.9 C throughout the entire incubation period.

Nine male and nine female chum salmon were used to test the possible effects of male size and age on alevin and fry growth. The fish were spawned so that three sets of a 3 x 3 factorial cross were created. Female size and age were kept constant in each factorial cross, whereas male size and age varied. Three males and three females were spawned at

the same time and the egg complement of every female was divided into thirds with each third being fertilized by a different male. In this manner, a total of 27 different crosses was created (Fig. 4).

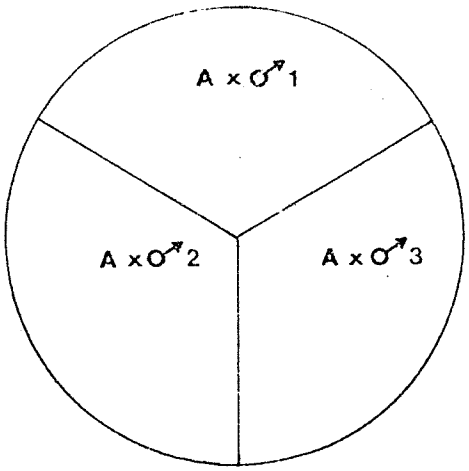
Each cross or population was kept separate by being incubated in its own Heath tray. Observations were made during the incubation period to determine the number of temperature units required to hatch eggs originating from the different crosses. The wet and dry weights of newly hatched alevins were also measured. Three groups of three alevins were removed from each cross, blotted dry, and weighed to the nearest .01 g on a Mettler P 120 balance. The groups were then placed in a drying oven and baked at 97° C for 24 hours. After being dried, the alevins were placed into a vacuum desiccator, allowed to cool, and reweighed on the Mettler P 120 balance.

A similar sample of nine fry/population was removed and weighed 102 days after fertilization. Also at this time, three groups of 100 fry from every cross were blotted dry on a damp sponge, placed in a tared beaker filled with water, and weighed to the nearest .01 g on a Mettler P 1200 balance. From these samples, 40 to 50 fork lengths (tip of snout to fork of tail) were obtained on fry representing each cross.

Immediately after sampling, the fry from each population were individually counted and transferred into rearing troughs. The troughs were 30.7 cm x 30.7 cm x 240 cm and divided by wooden-framed nylon-mesh (1/16-inch Delta) screens into three equal rearing areas 30.7 cm x 30.7 cm x 78.5 cm. During the 13-week rearing period, water depth was maintained at 16.5 cm, velocity at 11 liters/min (3 gal/min) and temperature varied between 8.7 C and 8.9 C. Dissolved oxygen concentrations in each trough were determined on a weekly basis by a Hach kit and were moderately low, ranging from 9.8 ppm to 5.4 ppm.

To test for possible location effects within the troughs, the three populations of fry produced from a male were placed into all three possible rearing areas (head, mid, and tail) in three different troughs, whereas the progeny originating from a single female were always reared in the same divided trough. Thus, each trough contained three populations of fry begot by the same female yet fertilized by different males (Fig. 5).

The fry were fed in excess 14 to 16 times/day on an Oregon Moist Pellet diet. During the period they were being reared, each population was sampled once a week. Twenty-five fry were randomly selected from each cross, killed with a heavy dosage of MS-222, and individual lengths (TSFT to the nearest .25 mm) and wet weights (to the nearest .001 g) were then taken. The sacrificed fry were next arbitrarily divided into five lots of five fry each and baked in a drying oven for 48 hours at 97 C. Upon removal from the oven, the fry were placed into a vacuum desiccator, allowed to cool, and weighed to the nearest .001 g on a Mettler P 120 balance.



THE EGG COMPLEMENT OF ♀ A

♀♀ \ ♂♂	1 (3yrs)	2 (4yrs)	3 (4yrs)
A (4yrs)	A x 1 (4 x 3)	A x 2 (4 x 4)	A x 3 (4 x 4)
B (4yrs)	B x 1 (4 x 3)	B x 2 (4 x 4)	B x 3 (4 x 4)
C (4yrs)	C x 1 (4 x 3)	C x 2 (4 x 4)	C x 3 (4 x 4)

A REPRESENTATIVE 3X3 FACTORIAL CROSS

Fig. 4. Three 3x3 factorial crosses were created by spawning three different groups of three males and females at the same time. In making the factorial crosses, the egg complements of every female were split into thirds, with each third being fertilized by a different male. In this fashion, 27 distinct crosses (15, 4-year-old females x 4-year-old males and 12, 4-year-old females x 3-year-old males) were produced.

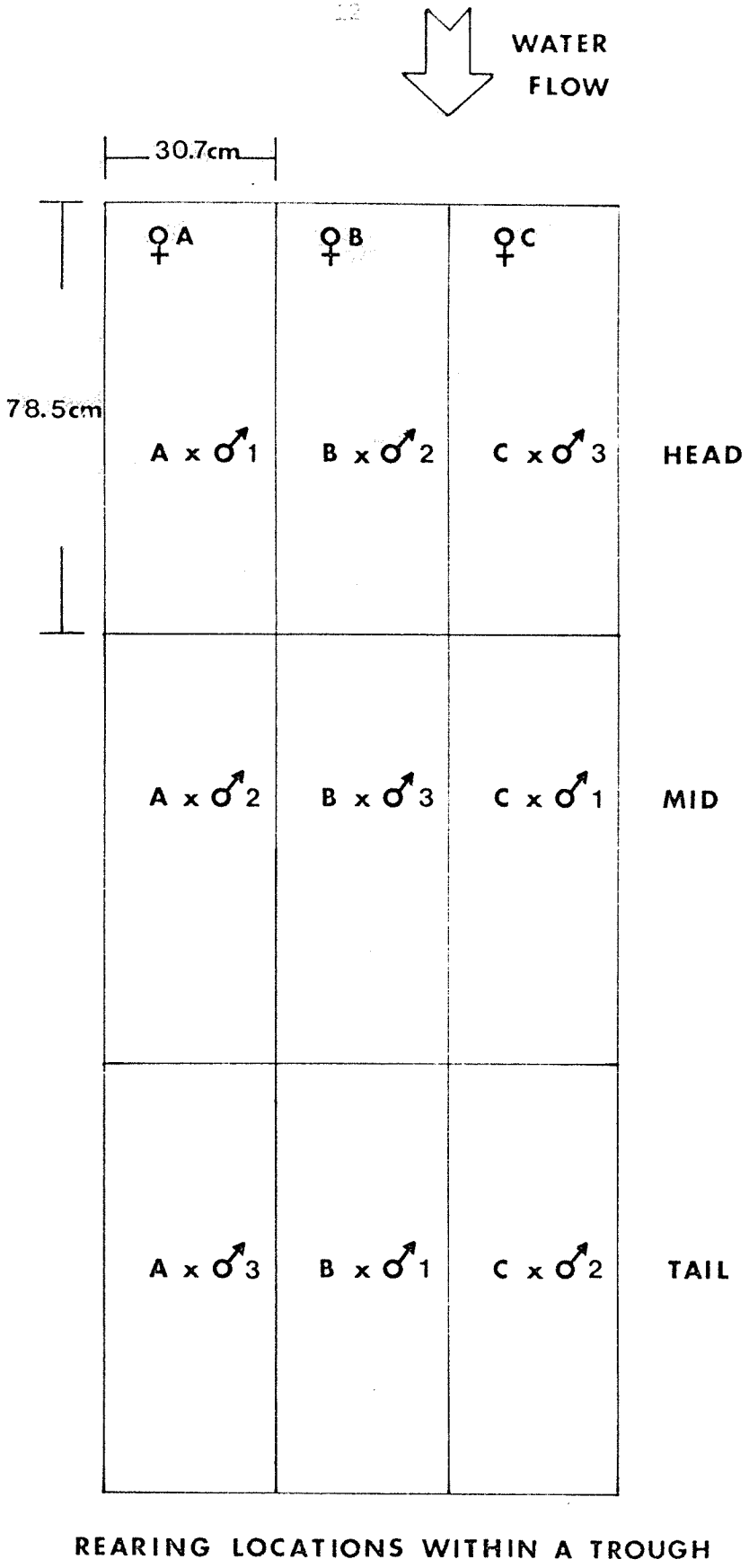


Fig. 5. The rearing locations within a trough for fry begot from a single 3x3 factorial cross.

Possible maternal effects on the developmental rate and size of fry at emergence were also examined. The eggs from three females (one 4-year-old and two 3-year-olds) were fertilized by two similarly sized 3-year-old males. A sample of eggs from each female was removed at the time of fertilization and allowed to water-harden for 24 hours. After water-hardening, the eggs were measured and weighed as previously described.

The remaining fertilized eggs from each female were placed in separate wire egg baskets and hung in rearing troughs. While incubating in the wire baskets, the eggs were kept in total darkness except during periods of routine sampling. Just before hatching, the eggs were removed from their baskets and transferred into Heath trays furnished with Astroturf substrate.

To determine if differences existed in the rate at which the eggs developed, three eggs from each lot were removed daily and preserved in 5% acetic acid. After the eggs hatched, alevins were removed once every 7 days and preserved in 5% Formalin. The yolk and larval tissue material from each alevin were separated and both parts were weighed, dried, and reweighed.

Results and Discussion

The morphometric properties of both the females and eggs originating from the middle run are presented in Tables 1 and 2. The tables are designed to allow comparisons between 3- and 4-year-old females which represent the dominant age classes returning to this stream. Not unexpectedly, 4-year-old females are larger and produce bigger eggs than 3-year-olds.

Linear regression analyses were used to test whether correlations existed between a female's age, weight, length, and condition factor and the diameter, weight, and condition factor of her eggs. These tests indicated that there was little linear correlation between the physical characteristics of 4-year-old females and their eggs. Yet, moderately strong correlations did exist between these characteristics (except female condition vs. egg condition) in 3-year-old fish.

Because the eggs produced from variously sized females originating from one population are subjected to similar selection pressures, it should not be too surprising that there are moderate or low correlations between a female's physical parameters and the size of her eggs.

Eggs originating from 4-year-old females appear to have a greater nutritive value than those produced by younger fish. Koski (1975) demonstrated that fry produced from 4-year-old females weighed more upon their emergence than those produced from 3-year-old fish even though the diameter of the eggs was the same. This result may have been caused by a difference in the nutrients within the eggs or the presence of a

Table 1. A comparison between the lengths, weights, condition factors[1], and egg parameters of 3- and 4-year-old females migrating into Big Beef Creek during November and December 1974

Age	Length		Weight		Condition factor		Egg diameter		Wet egg weight		Dry egg weight		Condition factor[2]	
	$\bar{x}$ value	SD	$\bar{x}$ value	SD	$\bar{x}$ value	SD	$\bar{x}$ value	SD	$\bar{x}$ value	SD	$\bar{x}$ value	SD	$\bar{x}$ value	SD
4	735 mm	29 mm	4.7 k	.6 k	1.17	.07	7.16 mm	.24 mm	.27 gr	.025 gr	.105 gr	.010 gr	.735	.046
3	664 mm	37 mm	3.4 k	.6 k	1.15	.12	6.72 mm	.22 mm	.23 gr	.021 gr	.088 gr	.088 gr	.742	.030

[1]Condition factor = (body weight/fork length^3) x 10^5.

[2]Egg condition factor = (wet egg weight/egg diameter^3 x 10^3.

SD = Standard deviation.

Table 2. Conditional[1] linear regression analyses between the morphometric characteristics of 3- and 4-year-old chum salmon females and their eggs

Age	X variable	Y variable	r	r^2	B value	t test B = 0	Significance > .05
4	wet egg weight	Body weight	.387	.150	9574	5.104	Yes
3	wet egg weight	Body weight	.693	.481	20421	6.461	Yes
4	dry egg weight	Body weight	.432	.187	26815	5.828	Yes
3	dry egg weight	Body weight	.723	.522	51892	7.016	Yes
4	wet egg weight	Fork length	.308	.095	357	3.943	Yes
3	wet egg weight	Fork length	.622	.387	1114	5.335	Yes
4	dry egg weight	Fork length	.367	.134	1063	4.793	Yes
3	dry egg weight	Fork length	.657	.432	2868	5.858	Yes
4	egg diameter	Body weight	.335	.112	864	4.325	Yes
3	egg diameter	Body weight	.724	.524	2003	7.045	Yes
4	egg diameter	Fork length	.276	.076	33	3.487	Yes
3	egg diameter	Fork length	.638	.408	107	5.564	Yes
4	egg condition[2]	Condition factor	.054	.003	.079	.663	Non-linear relationship
3	egg condition	Condition factor	.091	.008	.376	-.613	Non-linear relationship

[1]Linear regression analysis assumes that the independent variable X is fixed and measured without error. In the above analyses the independent variables were measured with error and hence the standard (Model I) tests for significance may not be applicable to this data unless we assume it is a typical Berkson case. The Berkson case assumes that the independent variables are measured with error but that the X values and the error terms associated with them are not correlated thus allowing one to use the so-called Model I regression methods for tests of significance (Sokal and Rohlf, 1969). Because these assumptions were made, the regression analyses have been labeled "conditional" to differentiate them from typical Model I or Model II regression analyses (McCaughran, personal communication).

[2]Egg Condition Factor = (Wet egg weight/egg diameter^3) X 10^3.

different relationship between egg weight and diameter in 3- and 4-year-old fish. Since strong linearity was shown to exist between egg diameter and dry weight (Table 3), it was possible to test with an analysis of covariance whether there were differences in the weights of eggs with similar diameters produced from differently aged females. This analysis indicated that the slopes but not the levels of the two regression lines were similar at the .05-level (Table 4). This implies that eggs from 4-year-old females are lighter at a given diameter than those originating from 3-year-olds.

To determine if a relationship existed between egg and fry weight, a linear regression analysis was completed that compared the dry weights of eggs and fry produced from nine 4-year-old females. The results of this analysis (Table 5) and data presented in Fig. 6 corroborate earlier findings by Koski (1975) that suggested a strong positive linear relationship between egg weight and fry size at emergence.

Parental influences on embryo growth rates and efficiency of yolk material utilization were also examined. It was found that egg weight did not influence the temperature unit requirements of eggs originating from 4-year-old females (analysis of variance $F = 2.88 < F\ .05\ [5,6] = 4.39$). However, it was discovered that eggs produced by 4-year-old females hatched sooner when they were fertilized by 3-year-old males than if by 4-year-old fish (analysis of variance $F = 4.8405 > F\ .05\ [1,25] = 4.24$). Withler and Morley (1970) examined the hatching times of interspecific hybrids made among sockeye (*O. nerka*), chum, and pink salmon (*O. gorbuscha*) and concluded that the female parent determined the embryo size at hatching, that the male parent controlled the rapidity at which the embryo reached this point, and therefore influenced the time at which the egg would hatch. It is speculated here that the inequalities observed in hatching times due to male age may reflect intrinsic differences in the metabolic characteristics of 4- and 3-year-old males.

Conversely, how efficient the embryo was in utilizing the energy resources of its egg was not influenced by the age of its male parent (analysis of variance $F = .01420528 < F\ .05\ [1,16] = 4.49$) nor apparently by egg size.

Fish Culture Results

Data gathered on the growth characteristics (weekly changes in dry weight) of reared fry produced from artificial crosses were analyzed by multiple regression analysis and analysis of covariance which incorporated dummy variables. First, the influence of seven variables (time reared, egg weight, age of male parent, rearing location within a trough, kg of fry/m^3, g of fry/liter per min, and absolute flow rate of water through a trough) on the dry weights of fry originating from each of the 27 experimental populations was determined by using multiple regression analysis. These variables accounted for 96% of the observed variance in the dry weight values of the examined fry.

Table 3. Conditional linear regression analyses between the diameter and dry weights of eggs begot by both 3- and 4-year-old chum salmon originating from the middle run of Big Beef Creek

Age	Relationship: X variable	Relationship: Y variable	Regression formula	r	r^2	F ratio: Value	F ratio: Significance .001	t test B = 0: Value	t test B = 0: Significance .001
3	dry egg wt	egg diam	$\hat{y}=4.733+22.619(x)$	.871	.759	141.9	Yes	11.912	Yes
4	dry egg wt	egg diam	$\hat{y}=5.288+17.875(x)$	.743	.552	182.7	Yes	13,516	Yes

Table 4. An analysis of covariance on the linear relationships observed between egg diameter and dry weight in 3- and 4-year-old chum salmon

Age group females	df	y^2	xy	x^2	df	Residuals s.s.	Residuals m.s.
4 years	149	8.7986	.2719	.0152	148	3.939	
3 years	46	2.2156	.0744	.0033	45	.534	
				Totals	193	4.473	.02318
		Difference for testing slopes			1	.061	.06054
	195	11.0142	.3463	.0185	194	4.534	.02337
		Difference for testing levels			1	.225	.22516
	196	17.8392	.6121	.0286	195	4.758	

For differences in slope $F = 2.6120 < F_{.05}(1,193) = 3.92$ therefore fail to reject the null hypothesis at the .05 level

For differences in level $F = 9.6357 > F_{.05}(1,194) = 3.92$ therefore reject the null hypothesis at the .05 level.

Table 5. A conditional linear regression analysis between the dry egg weights of nine 4-year old females and dry weights of their 102-day-old alevins

Relationship				F ratio		t test B = 0	
x variable	y variable	Regression formula	r	Value	Significance >.001	Value	Significance >.001
Dry egg wt	dry alevin wt	$\hat{y} = .0305 + .3938\ (x)$	.724	27.47	Yes	5.241	Yes

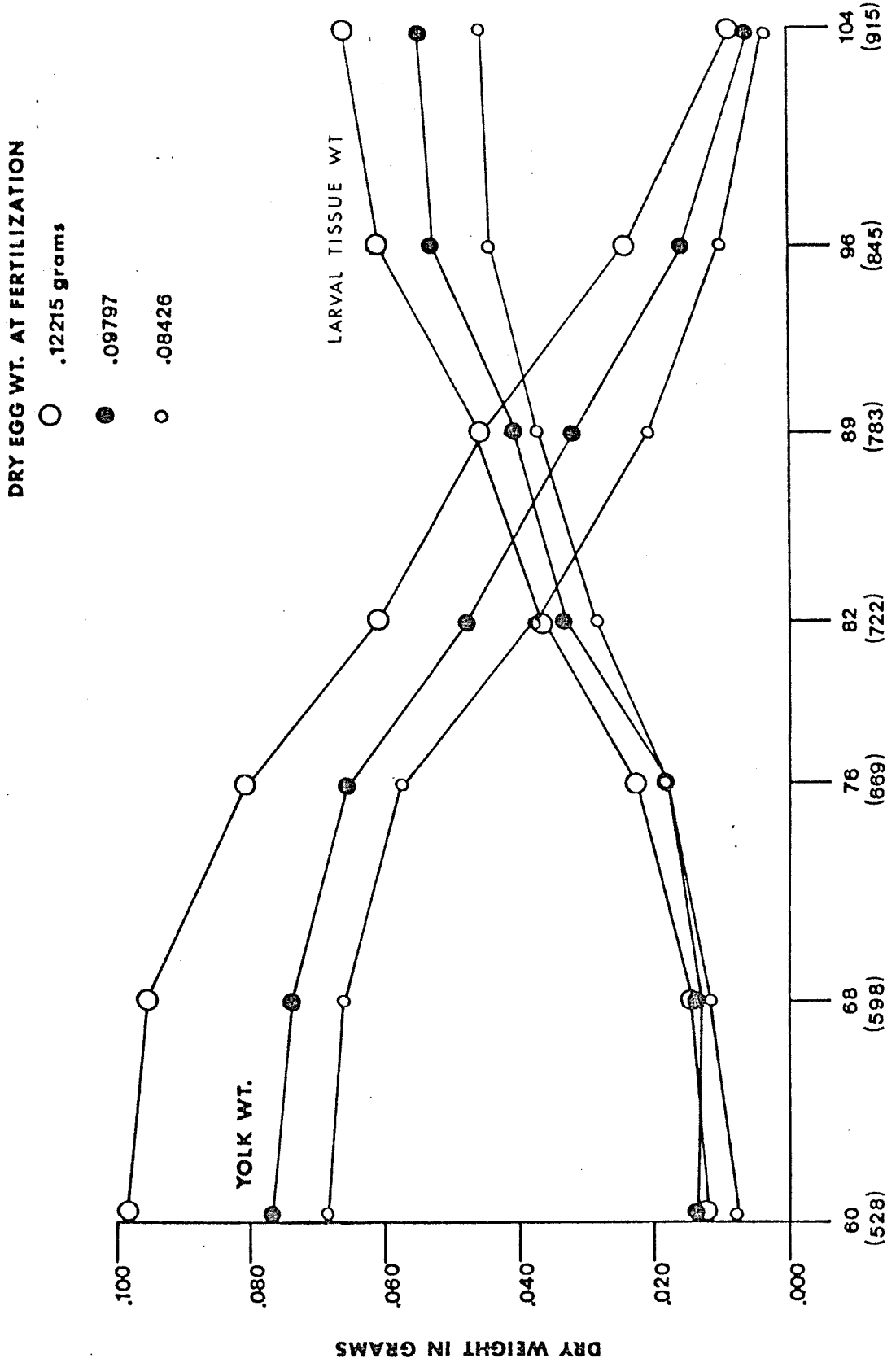


Fig. 6. Temporal changes in the dry weights of both yolk and somite materials in alevins produced from three batches of differently-sized eggs.

To determine if any of the variables could be eliminated from further analysis, the explained sum of squares was divided into components attributable to each independent variable. This was done by using the standard regression method where each variable is treated as if it had been added to the regression equation in a separate step after all the other variables had been included (Nie, et al 1975). Results in Table 6 illustrate the calculated F ratios used to test the significance of the regression coefficients of each variable used in the equation. This analysis indicated that the absolute flow rates of water within the rearing troughs had little effect on dry weight values and consequently, they were excluded from further analysis.

Second, by using analysis of covariance with dummy variables, it is possible to determine whether a regression model with several lines will reduce the variability of Y to a greater extent than one with only a single line. This is determined by evaluating the F ratio obtained from such an analysis. This technique was used to test whether egg size, male age, and rearing position within a trough influenced dry weight values of sampled fry. We found that 3-year-old males significantly increased the growth rates of the fry they fathered, ($F = 4.4 > F\ .001\ [5, 1828] = 4.10$), and that larger eggs produced larger fry ($F = 15.2521 > F\ .001\ [40, 1782] = 1.84$), and that rearing position[1] also influenced fry growth ($F = 198.7619 > F\ .001\ [10, 1818] = 2.96$).

Whether these differences can be attributed to genetic factors related to age at maturity is impossible to determine. We have no data on the genetic background of the parental fish other than that they originated from Big Beef Creek. To fully assess the impact of parental age (particularly male age) on fry growth would require the capture and crossing of adults with known genetic background. Until we can do this, we can only be tantalized by a myriad of questions which deal with the heritability of life-history strategies.

No matter how, or why, we make our parental crosses, we are still confronted with providing the resulting eggs and alevins with an optimum incubation site. The next section describes some experiments we carried out in Heath Tecna trays in an attempt to determine the effects of various incubation substrates on alevin survival and growth.

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[1]During a portion of the rearing period water was recycled back into the rearing troughs. While this occurred fry which were reared in the anterior or head portion of a raceway (see Fig. 5) experienced a greater mortality rate than those reared in a mid or tail portion. Excess food recycled back into the troughs apparently caused abrasions on the gills and other body surfaces which were later infected with Saprolegnia. Consequently the density of fry in a head portion of a raceway was reduced and this caused an apparent increase in the rate of growth on the surviving fish.

Table 6. Results of a stepwise regression analysis on chum salmon fry rearing data

Multiple Regression
Dependent variable: Log 10 transformation of the dry weights of sampled fry

Step	Variable	Simple r	F to enter or remove	Significance
1	Time reared	.972	35163.82	.001
2	Dry egg weight	.099	397.22	.001
3	Fry wt/liter/min	.573	170.32	.001
4	Male age	-.023	38.49	.001
5	Fry wt/m^3	.804	9.82	.002
6	Rearing location	-.024	4.99	.026
7	Absolute flow rate	.457	.72	.396

III. EFFECTS OF VARIOUS TYPES OF SUBSTRATES ON THE EFFICIENCY OF YOLK MATERIAL UTILIZATION IN CHUM SALMON LARVAE INCUBATED WITHIN HEATH TECNA INCUBATORS

Introduction

Artificial propagation of chum salmon in hatcheries has not generally enjoyed a great deal of success in North America. This has been attributed to the presence of incompatible incubation environments that often exist in such facilities (Brannon, 1965; Bams, 1967 and 1969; Bailey and Heard, 1973; and McNeil, 1973). Clearly, to optimize the potential of each cultivated egg, it is necessary to design and create incubation environments that allow the egg and alevin to develop naturally and consequently, to utilize its yolk materials in the most efficient way possible. This need has been recognized by quite a few investigators (Disler, 1953; Brannon, 1965; Bams, 1967 and 1969; Poon, 1970; Emadi, 1972; McNeil, 1973; and McNeil and Bailey, 1975) and provided for in chum salmon hatcheries in Russia and Japan (McNeil, 1973; and Mathews and Senn, 1975).

Recently, Bob Dewey (personal communication) had the ingenious idea of utilizing plastic Astroturf as an artificial incubation substrate for incubating sockeye eggs and larvae. Bailey, Taylor, and Pella (1975) used the same incubation material and developed experiments that examined the effects of this substrate on pink salmon eggs. Their studies broached a very important question by examining the relationships existing among waterflow, fry density, incubation substrate, and fry size at emergence in artificial incubation structures. As a result of these studies, everyone involved in similar work is carrying a different form of plastic in his pocket. Yet, little work has been done in evaluating the efficiency of yolk material utilization on these substrates. We had the opportunity this past year to evaluate how efficient chum salmon alevins were in utilizing their yolk materials while incubating in Heath Tecna incubator trays equipped with either a gravel, Astroturf, or bare screen substrate.

Methods, Materials, and Results

Eyed eggs from ten 4-year-old females (Table 7) were randomly mixed and placed into Heath Tecna incubator trays at a density of three alevins/cm^2 (20 alevins/square inch). The incubators were supplied with water from an artesian well that ranged in temperature from 8.7 C to 8.9 C throughout the entire incubation period. After 99 days (866 TU's C), the incubation period was completed and five samples of 100 fry from each treatment group were sampled gravimetrically to calculate the average wet weight of the fry. Additionally, to determine average lengths, 30 fry from each treatment were measured (TSFT) to the nearest mm. Significant weight and length differences of approximately .1 g and 3 mm occurred among the groups (Table 8). Average dry weights of fry

Table 7. Average adult size and egg data parameters for the 10 chum salmon females used in the substrate experiments at Big Beef Creek, 1974

Date spawned	Age	$\bar{X}$ weight (g)	$\bar{X}$ length (mm)	$\bar{X}$ egg diameter (mm)	$\bar{X}$ egg weight wet (g)	dry (g)
12/8/74	4	4765	746	6.934	.258	.102

originating from each treatment were determined and used in conjunction with dry weights to compute how efficient the alevins had been in converting yolk materials to larval tissue. Fry incubating on screen substrates proved to be 20% less efficient in utilizing their yolk materials than those incubating on either gravel or Astroturf (Table 9).

A one-way analysis of variance, using the wet weights of fry incubated on the various substrates was performed (Table 10) and showed that a highly significant ($P < .005$) difference existed among fry originating from the substrate treatments. A Tukey's w-procedure (honestly significant difference test) performed on the same data (Table 11) indicated that fry incubated on bare screen were different (at the $P < .01$ level) from those which had been placed into trays with Astroturf or gravel.

To determine if this difference in size would persist over time, fry from each treatment were reared in freshwater under identical conditions. The growth rates of all the groups were similar, but because of their initially smaller size, fry originating from trays with bare screen were never able to catch up in weight or length to the other two groups (Fig. 7).

Discussion

Astroturf appears to provide an excellent substrate for developing chum salmon larvae when they are incubated at low hatching densities (≤ 3 alevins/cm^2) with adequate water exchange (11 liters/min). Its use in incubation systems relying on groundwater sources which have periodic sediment loads has yet to be examined. Mortalities can be difficult to remove from this type of material and mats of fungus may develop which could significantly reduce egg-to-fry survival.

The yolk material utilization of chum salmon larvae originating from large eggs was slightly reduced when they were incubated on Astroturf with bent blades. Table 12 shows the results of a linear regression analysis between the dry egg weights of nine 4-year-old females and the efficiency of their embryos in utilizing yolk material. The observed relationship may be due to the inability of larger alevins to successfully penetrate into the plastic grass and, hence, they may spend more time swimming on the surface of the substrate than alevins produced from smaller eggs. Finally, Astroturf is difficult to attach to Heath trays and, as a result, may not be economical for large-scale use in production hatcheries.

These disadvantages have led us to test the suitability of different forms of Astroturf and other plastic devices as incubation substrates for developing chum salmon.

Table 8. The average wet weight (five groups of 100 fry from each treatment) and length (30 measurement/treatment group) of chum salmon fry incubated on different substrates in Heath Tecna trays

Physical parameters	Unaltered Heath screen tray	Treatment: Heath tray with Astroturf mat	Heath tray with gravel substrate
Average wet weight/ fry (g)	.2843	.3838	.3820
Average length (mm)	33.96	36.76	37.38

Table 9. The average dry weight (5 groups of 3 fry each) and percentage of yolk material utilization of fry originating from each treatment group

Physical parameters	Unaltered Heath screen tray	Treatment: Heath tray with Astroturf mat	Heath tray with gravel substrate
Average dry weight/fry (g)	.0515	.0734	.0745
Average % of yolk material converted to larval tissue	50.72	72.36	73.43

Table 10. Analysis of variance on the wet weights of chum salmon fry incubated on three different substrates in Heath incubator trays

Source of variation	SS	d.f.	Ms	F
Between locations	324.21	2	162.11	365.4
Within locations	3.44	12	0.286	
Total	327.65	14		

$F = 565.4 > F_{.005}(2, 12) = 8.51$ reject the $H: T_i = 0$

Table 11. A comparison of all treatment (substrate types) mean wet weights by Tukey's w-procedure (honestly significant difference test)

Treatment	Heath tray with gravel substrate	Heath tray with Astroturf mat
Unaltered Heath screen tray	9.95[1]	9.77[1]
Heath tray with Astroturf mat	0.178	

[1]Values > W .01 level (1.206) indicate differences between the mean wet weights of fry produced from the various substrate types

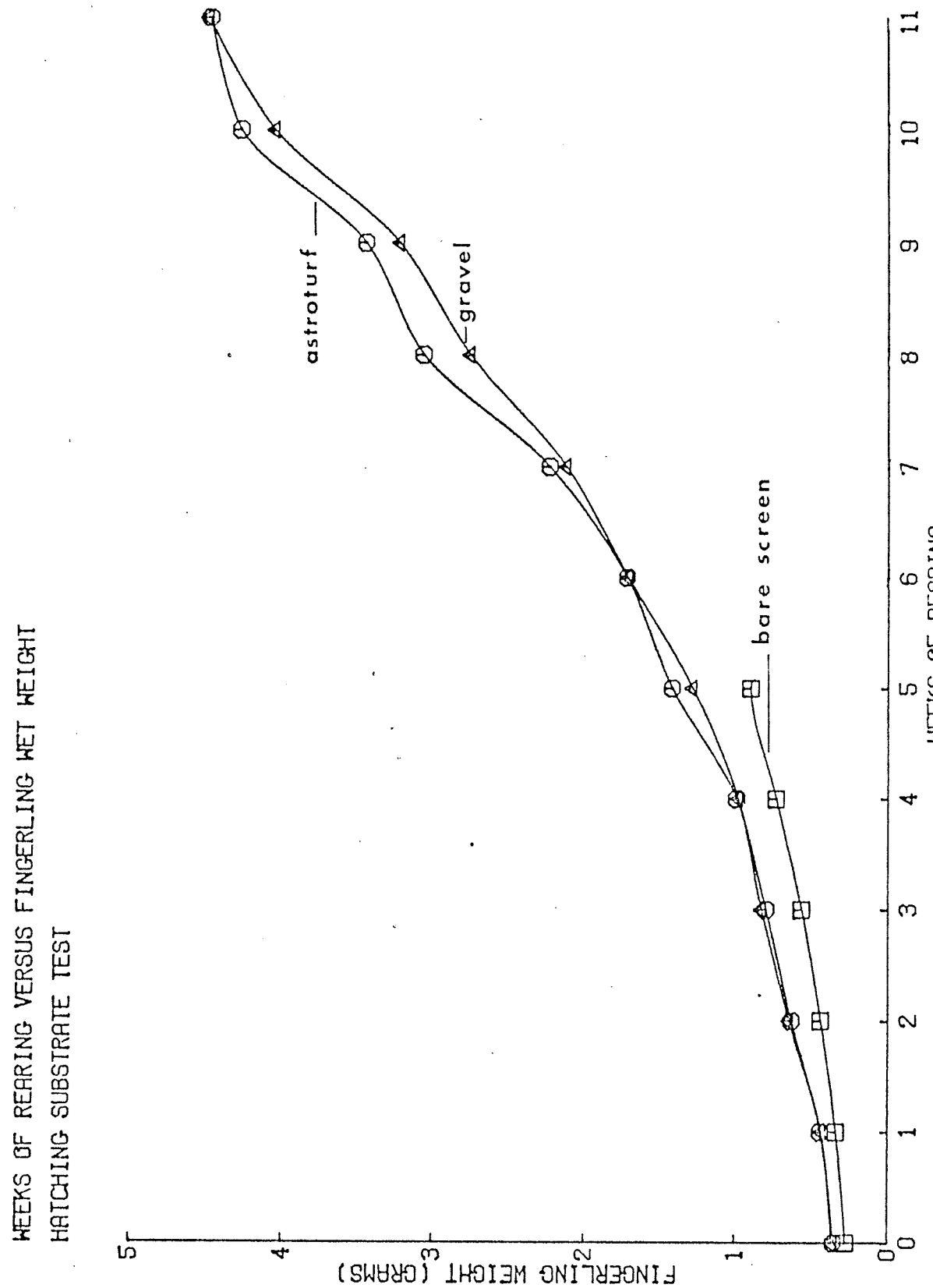


Fig. 7. The growth rates of chum salmon fry incubated within Heath trays equipped with either a gravel, screen, or Astroturf substrate.

Table 12. A linear regression analysis on the relationship between dry egg weight and embryo efficiency in the utilization of yolk materials as exhibited by the progeny produced from nine 4-year-old female chum salmon

Relationship		Regression formula	r	F ratio		t test B = 0	
X variable	Y variable			Value	Significance >.001	Value	Significance >.001
dry egg weight	% of dry egg weight converted to dry alevin weight	y = 98.25-282.04(x)	-.595	13.928	Yes	3.732	Yes

1975-1976 Incubation Substrate Experiments

This year, one of us (Bruce Snyder) is examining the effects of four substrate types (bare screen, gravel, various types of Astroturf, and biofilters) on chum salmon alevins at different loading densities in hatcheries with and without sediment-free water. The densities being examined are 2, 2.5, 4.3, and 6 alevins/cm^2 (13, 15, 27, and 38 alevins/$inch^2$). A constant flow of 18 liters/min (5 gal/min) is being provided to each Heath incubator tray used in these experiments.

The most promising new substrate being tested this field season appears to be the Actifil biofilter (bio-ring) manufactured by the Norton Chemical Process Products Division, Akron, Ohio. Biofilters come in a variety of sizes and weights and we are testing only the 1-, 1.5-, and 2-inch floating and sinking types this year. The biofilters appear to stop unnecessary fry movement quite well at the loading densities we are examining.

IV. DEVELOPMENT OF A PROTOTYPE INCUBATION BOX FOR CHUM SALMON EGGS AND LARVAE

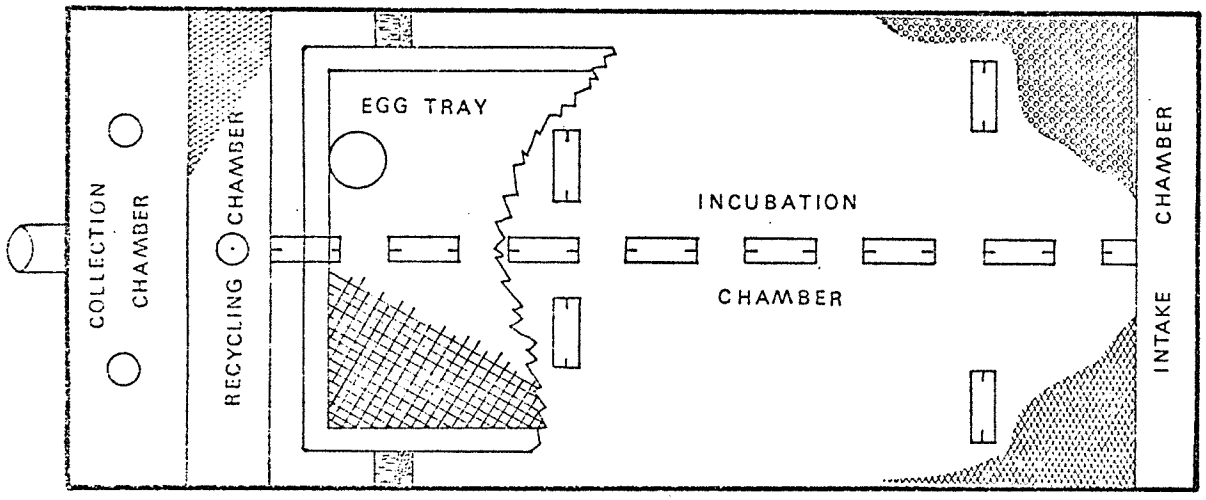
Introduction

Besides using Heath Tecna incubators to test the effects of various substrates on chum salmon larvae, we also designed and used a small portable egg-incubation box for similar experiments. A variety of such boxes has previously been developed (Bailey and Heard, 1973; Bams, 1973 and 1974; Bailey and Taylor, 1974; Wilson, 1974; and Lannon, 1975) and they have used either stream or crushed gravel as incubation substrates. These boxes have all successfully produced unfed Pacific salmon fry. One common complaint, however, has been the necessity to remove and clean the incubation gravels. The use of plastic substrates, such as Astroturf or Biofilters, may provide a solution to this problem. These materials are reusable and light; they are also easily cleaned, disinfected, and re-installed.

The Design and Use of the Snyder Incubation Box

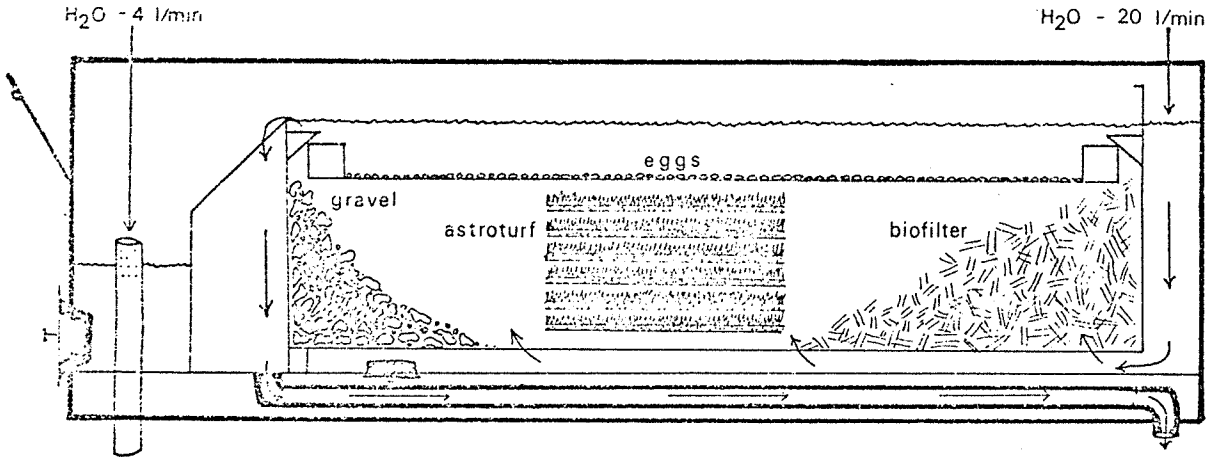
The sides and bottom of the incubation boxes (123 cm x 50 cm x 390 cm) were made from exterior 1.9-cm (3/4-inch) plywood, and given a coat of Fiberglas resin. A portion of each box (the incubation chamber) was equipped with a raised (2.5-cm) false bottom (46-cm x 92-cm) of 1.5-mm^2 plastic screening. To permit maximal use of space and water, the boxes were equipped with flanges which allowed them to be stacked.

Internally, each box (Fig. 8) is partitioned into three chambers. The intake chamber directs incoming water to a baffle which creates an upwelling flow in the incubation chamber. To ensure that the flow is

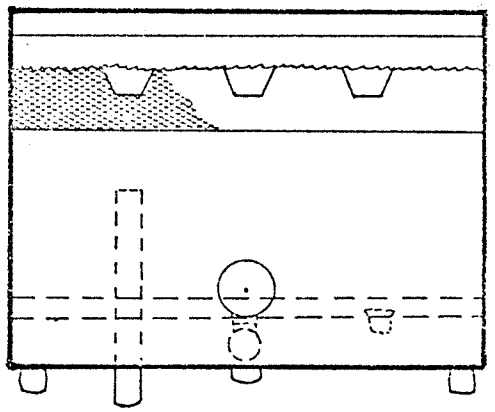


TOP VIEW

	WIDTH (cm)	LENGTH (cm)	HEIGHT (cm)	VOL (cm^3)	AREA (cm^2)
INTAKE CHAMBER	5.5	42	27		
INCUBATION CHAMBER (Maximum substrate depth)	46	92	18	76,176	4232
RECYCLING CHAMBER	5	42	18		
COLLECTION CHAMBER	10	42	18	7,560	420
EGG TRAY (Capacity: approximately 30,000 eggs)	34	80	3		2636



CROSS-SECTIONAL VIEW



FRONT VIEW

Fig. 8. Schematic views and specifications for the egg incubation box developed and used at Big Beef Creek.

evenly distributed through the plastic screen bottom of the incubation chamber, a series of small wooden baffles was glued to the bottom of the box.

The third chamber is subdivided into water recycling and fry collection chambers. The entrance of the recycling portion of the chamber is covered with 1.5-mm^2 plastic screen and slopes with a 45-degree angle toward the fry collection chamber. As water leaves the incubation chamber, it passes through the screen and is led into a 2.5-cm, -diameter pipe, which runs back underneath the box and empties into the intake chamber of the box lying directly beneath it. The screen helps aerate the water as it circulates through a stack of boxes, and passes emerging fry originating from each box into their respective collection chamber.

None of the water flowing out of the incubation chamber enters the collection chamber. Instead, the uppermost collection chamber in a stack of boxes is provided with water from a separate (4 liter/min) source. Water depth in each collection chamber is regulated by a perforated standpipe that can be raised or lowered. The standpipes are staggered within a stack of boxes by anchoring them into one of two 2.5-cm diameter holes located on the bottom of the chamber. A rubber stopcock is placed into the hole which is not in use. Water is recycled from one collection chamber to the next by flowing through the standpipes into the chamber directly beneath it.

Fry can be removed from each chamber either by siphon or out an exit drain (5-cm diam) located on the front end of the box (Fig. 8). The collection chamber can be inspected during fry incubation and emergence by using a hinged door which is also located on the front end. With some additional plumbing, fry originating from a stack of boxes can be funneled through their respective exit drains into one or more troughs. When not in use, the drains are closed with expandable rubber stoppers. The rest of the box can be drained by removing a similar plug located on the bottom of the incubation chamber.

To fill the boxes with either green or eyed eggs, wooden framed trays (92 cm x 44 cm) with 6.3-mm nylon mesh net bottoms are used. A single tray loaded with eggs is locked over the (96-cm x 46-cm x 21.5-cm) incubation chamber and not removed until hatching is completed. After hatching, the alevins fall through the netting and soon bury themselves in the offered substrate (up to 15 cm deep). While in use, the tops of the uppermost boxes in a stack are covered with black plastic to prevent light from penetrating into any of the incubators.

Twelve incubation boxes were built and supplied with 17 liters/min of artesian (8.7 C to 8.9 C) water. Nine of the boxes were used to examine the effects of substrate type (bare screen, stream gravel 12 cm to 28 cm in diameter, and Astroturf), substrate depth (2.5 cm to 5 cm), and alevin density (3.1, 3.9, or 4.7 alevins/cm^2) on survival and yolk utilization. Each substrate type was tested in three boxes with each having a different density of alevins. Moreover, the boxes containing Astroturf and gravel were supplied with different depths of substrate material.

Results and Discussion

The incubation boxes are designed so that emerging fry can leave the incubation chamber on their own volition. Consequently, the numbers of fry emerging from each box were determined daily by counting the fry in the collection chambers. Premature emergence of fry occurred in all the boxes. Small numbers (< 100) of alevins were observed entering the collection chambers soon after hatching (435 to 766 temperature units C). As the alevins became more developed, greater numbers of them left their respective incubation chambers and in some cases, high mortalities occurred in the collection chambers due to suffocation. Because the number of premature alevins originating from each box was large, it was impossible to make comparisons among the treatment groups regarding the effects of alevin density or substrate type on fry quality. However, both the gravel and Astroturf substrates proved to be more successful in retaining developing alevins within their incubation chambers than those with only plastic screen bottoms.

These results meant that modifications in the box were necessary before it could be used as an effective research or enhancement tool. An increase in substrate depth, the use of laminar flows, or the addition of a removable screen lid over the incubation chamber may be all that is required. This past fall (1975), the same boxes were used again, except that the substrate depth was increased to 15 cm. The increase in substrate depth proved to be very effective in reducing premature emergence of alevins, and further refinements in the box may not be necessary.

V. TRIALS AND TRIBULATIONS OF CHUM SALMON CULTURE IN THE HATCHERY AND IN REARING POOLS

Introduction

Recently, Kobayashi (1976) has shown that a spectacular increase (≃ 1.1%) in fry-to-adult survival occurs in Asian chum salmon stocks if the young are first reared in freshwater and then released at an "appropriate" time. Lannon (personal communication) feels that freshwater rearing of North American chum stocks should prove just as successful. The success of the Japanese chum salmon enhancement program has re-emphasized the importance of understanding the selection pressures an enhanced stock experiences.

Before an extensive rearing program can begin, a considerable number of questions need to be answered. These include concerns about incubation and rearing facilities, diet, disease, rearing densities, effects of water temperature, genetic background on growth, and when to release the reared fry.

We attempted to examine some of these questions and this section will briefly describe: 1) our efforts to release fed chum salmon fry representing four distinct parental crosses based upon age, and 2) the facilities, diet, disease problems and treatment, and the incubation and rearing densities we used.

Procedures and Materials

A minimum of 50,000 fingerlings from each parental cross (3-yr-♀ x 3-yr-♂, 3-yr-♀ x 4-yr-♂, 4-yr-♀ x 3-yr-♂, and 4-yr-♀ x 4-yr-♂) was to be marked and released to evaluate the survival rate and predominant age of return of the progeny produced from each cross. Two-hundred seven (47, 3-yr-olds and 174, 4-yr-olds) females were spawned as previously described so that fecundity and other egg data information could be obtained. Spawning took place during the peak of the middle run (Nov. 20 to Dec. 11) to insure the availability of fish representing the desired age classes.

After fertilization, the eggs from three to four females representing the same parental cross were combined and incubated in standard Heath incubator trays until closure of the blastopore. During this developmental period (≃ 40 days) the eggs were picked 24 hrs after fertilization and not handled again. Just prior to hatching, the eggs were shocked and any unfertilized or aborted eggs were removed.

The eyed eggs were then transferred into Heath trays which were equipped with an Astroturf mat. The (1,340 cm^2) mats were attached to the bottom screen of the trays by using a Dennison Buttoneer and either plastic or Neoprene buttons. A standard density of 3 alevins/cm^2 (McNeil, personal communication) was predominately used and determined gravimetrically by weighing out an appropriate amount of eggs from each lot. Other densities of alevins were also examined to help determine whether differences in yolk material utilization would occur if the density of embryos within an incubator tray varied. All of the 44 densities examined were established gravimetrically and ranged from < 1.5, 1.6 to 2.2, 2.5 to 2.9, 3.0 to 3.5, and 3.8 to 4.3 alevins/cm^2. After 875 (C) temperature units had elapsed, the mean wet weight and length of fry from each population was calculated.

Water flow was kept at 11 liters/min while the eggs were reaching the eyed stage and increased to 15 liters/min thereafter. The developmental progress of the fry in each tray was routinely monitored and any dead eggs were removed to prevent fungus mats from developing in the Astroturf. Consequently it was not necessary to treat any of the trays with malachite green to control fungus growth. Moreover, during the incubation period each stack of incubator trays was covered with black plastic to eliminate the effects of light on the developing embryos.

The fry from each parental cross were removed from their respective trays after acquiring 875 temperature units. At this stage of development, the fish were almost "buttoned" having only a small slit on the abdomen. Survival and average weight of fry in each incubator tray were determined by weighing 3 groups of 100 fry plus all the remaining fish. These weights were incorporated into a simple algebraic formula and used to estimate the number of fry originating from each tray. Average lengths (TSFT) were also obtained by measuring a minimum of 30 fry from each tray.

As soon as the fry were processed out of the hatchery they were to be transferred to the Big Beef Creek Fish Research Station and reared there in portable swimming pools. However, water temperatures at this site were quite low and consequently the fry were initially reared at the hatchery. Since the artesian water supply utilized by the hatchery was not abundant enough to permit simultaneous incubation and rearing programs, an electric pump was installed to recycle a portion of the discharge water for the rearing program.

After an initial rearing period of 2 to 3 days in deep troughs (2.5-m x 0.5-m x 0.5-m) the fry were moved into 1.2-m x 1.84-m x 1.2-m nylon mesh (3-mm) pens which were placed in earthen ponds adjacent to the hatchery. When the water temperature in Big Beef Creek equaled that of the hatchery the fry populations were moved to this site and reared in portable, circular swimming pools (4.6-m to 5.5-m in diameter). Center drains were built into the pools and allowed them to be partially self-cleaning. Artesian well water was to have been used in the pools (189 liters/min per pool) but this was unavailable and water from Big Beef Creek was used instead.

Each population was fed 4 to 6 times daily with Oregon Moist Pellets and routine samples for length (TSFT) and wet weight were made every 2 weeks. Average wet weights were determined by randomly collecting 3 groups of 100 fry and weighing each group in a tared beaker filled with water on a Mettler P1200 balance. Length measurements were taken from 30 individuals randomly chosen from the previously weighed samples.

The fry were reared until they weighed from 2.3 to 4.5 grams each. At this time each population was to be marked by clipping different combinations of the dorsal and ventral fins and then released into the estuary of Big Beef Creek. The fry-to-adult survival rate, and predominant age of return for each parental cross was to be calculated by observing the number of adults bearing each mark.

Results and Discussion

Egg Collection, Fertility, and Incubation Mortalities

Four distinct populations (based upon parental age) were created from 691,100 eggs. Table 13 shows how the eggs (142,100 from 3-yr-old females and 549,000 from 4-yr-olds) were allocated to produce each population. Overall fertility amounted to 96.3% and was evaluated after 352 temperature units(C) had elapsed.

The embryonic ontongeny of the eggs and alevins was observed and arbitrarily broken into phases of development that corresponded to conspicious morphological or behavioral stages. Table 14 lists the temperature units (C) and number of days required for embyros to reach each stage. Unfortunately, mortalities were high (20%) after hatching and were mainly caused by suffocation and handling of fry prematurely emerging from the experimental egg boxes. In comparison, fry which were incubated in modified (attached Astroturf substrate) Heath Tecna trays experienced $\leq$ 1% mortality at the same stage of development.

A wide array of alevin densities (< 1.5 to 4 alevins/cm^2) were utilized in the modified Heath incubator trays. Since the alevins were placed into similar incubation environments (identical resting substrates and equivalent water flows), it was possible to examine the effects of crowding on the ability of the alevins to utilize their yolk materials. This was done by performing a conditional linear regression analysis between wet weight after water absorption and wet alevin weight after 875 temperature units (C) had been accrued. This analysis was performed on a number of populations and as Table 15 and Fig. 9 illustrate, a similar positive correlation between these variables exists in all the densities we examined. However, since we used sediment-free artesian water and never exceeded 3.1 alevins/cm^2 per tray in an entire incubator stack (16 trays), we do not know what the effects of uniformly high alevin densities (possible oxygen depression) and sediment-loaded water would have on these results.

Mechanical and Disease Problems Experienced During the Fry Rearing Program

After the incubation period was completed, fry from each population were placed into rearing troughs (1.1 kilo of fry/3.8 liters per min per trough) located inside the hatchery. For a short period (1 to 4 days) the fish were fed a fine, flour-like starter mash. This food caused abrasions on the gills which later became the sites of *Saprolegnia* infestation. The occurrence of this fungus caused a 2 to 3% mortality to occur. To reduce this mortality rate we recommend that crushed 1/32nd inch Oregon Moist Pellets be used as a starting diet rather than

Table 13. The allocation of collected chum salmon eggs into 4 populations based upon parental age

Type of parental cross Female x male Age of parental fish	Number of females used	Number of eggs allocated to each population
3 x 3	28	77,200
3 x 4	21	64,900
4 x 3	91	329,400
4 x 4	64	219,600
Totals	204	691,100

Table 14. Rate of embryonic development at a constant temperature (8.8C) for the middle run of chum salmon originating from Big Beef Creek

State of development	Time (in days) after ferilization	Temperature Units (C)
Closing of the blastopore	17	147.9
Beginning of eye pigmentation	25	217.5
Beginning of embryo movement	33	287.0
Hatching	58 to 61	504.6 to 530.7
Buttoning	109	948.3

Table 15. The relationship between wet egg weight and wet alevin weight at emergence for chum salmon fry incubated in Heath trays with Astroturf substrates at various densities

Alevins/cm^2	Number of trays	r	Regression formula
1.5	9	.884	y = 37.58 + 108.36 [X(I) - .265]
1.6 to 2.2	6	.924	y = 37.93 + 121.25 [X(I) - .265]
2.5 to 2.9	9	.976	y = 37.99 + 101.72 (X(I) - .271]
3.0 to 3.5	15	.854	y = 37.48 + 89.20 [X(I) - .260]
3.8 to 4.3	5	.999	y = 37.50 + 116.88 (X(I) - .242]
Mean for all densities	44	.909	y = 37.56 + 100.03 [X(I) - .262]

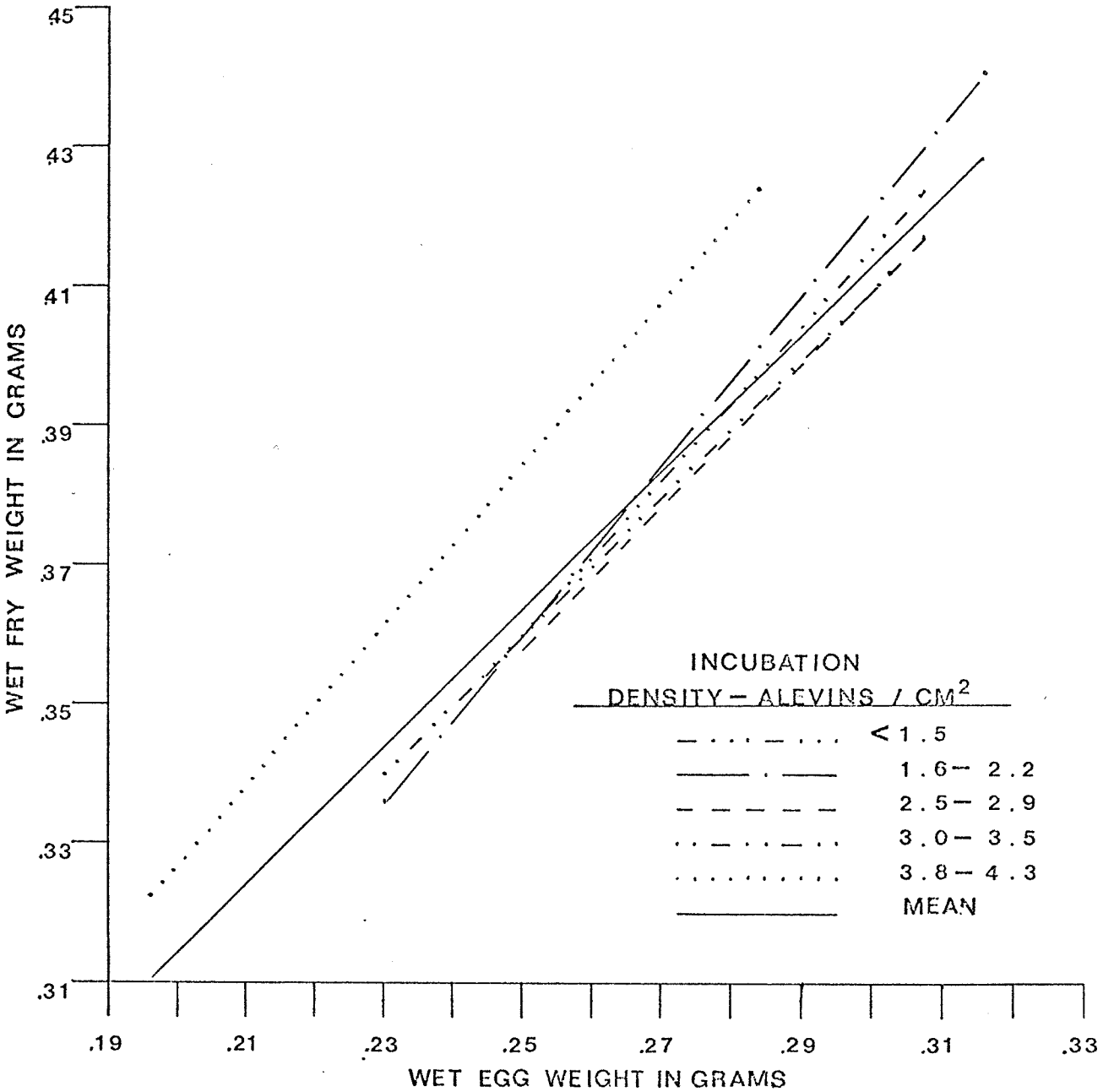


Fig. 9. The linear relationships between wet egg weight and wet alevin weight at emergence for chum salmon alevins incubated at various densities in Heath incubator trays equipped with Astroturf substrates.

the more traditional and abrasive starter mash. We tried this technique in 1976 and it worked very well.

Once the fry began to accept food they were transferred from the troughs and moved into pens located in earthen ponds adjacent to the hatchery. They were reared here at densities of 0.300 to 0.567 kilos of fish/0.027 m^3 until mid-April when they were moved to rearing facilities located at Big Beef Creek.

The two populations originating from 4-yr-old females (4 ♀ x 4 ♂ and 4 ♀ x 3 ♂) were placed into newly assembled, circular, portable swimming pools at an initial density of 0.473 kilos of fish/liter per min. The remaining two populations (3 ♀ x 4 ♂ and 3 ♀ x 3 ♂) were held in wooden raceways (0.9-M x 0.45-M x 5-M, 2.03 m^3) for three weeks at a final density of 1.05 kilos of fish/liter per min before being placed into similar pools.

The collapse of the pool containing the 4 ♀ x 3 o population (173,500 fry) caused a 60% mortality to occur in this group and disrupted the entire rearing schedule. Other factors besides mechanical failure also caused additional mortalitites. From May 10 to 20, losses in the pools increased to 2%/day from what appeared to be a fungal growth on the gills. Diquat bath treatments were given, but the cause was later identified on May 21 as a furunculosis bacteria, *Aeromonas salmonicidia*. The strain we were dealing with was highly virulent and resistant to terramycin, sulfamethazine and orthromycin. Furox 50 (furazolidone) was found to be effective and was incorporated into the diet until the fish were released on June 4th. Coho salmon smolts native to Big Beef Creek, which were being held temporarily for tagging studies by the Washington Department of Fisheries, proved to be the source of the furunculosis bacteria (cultures of the furunculosis bacteria were obtained from the coho smolts and identified by Drs. R. Antipa and D. Amend and verified by Mr. J. Wood).

Besides the furunculosis outbreak, otter predation and possibly heavy siltation also caused significant loses. On May 21 some unauthorized road construction on a small tributary one mile above the rearing site caused heavy siltation throughout the lower mile of the stream. Mortalities in the pools immediately doubled and attempted drug therapy was ineffective thereafter.

In summary, the four populations experienced an overall mortality rate of 92.3%. Over 50% of the mortalities could be attributed to furunculosis, while the remaining 40% were caused by mechanical failures, handling stress, otter predation and possibly heavy siltation. The surviving fry were released at the mouth of Big Beef Creek. Because the number of fry representing each cross was unequal and small, none of the groups was marked.

Growth Rates Experienced by the Reared Population

Meaningful comparisons among the growth rates of fry representing the 4 parental crosses (see Table 13) were not possible for a variety of reasons. First, each population (fry from each type of parental cross) experienced a unique rearing environment (different flow rates, population density, and water temperature regime) and secondly, since the populations were created as parental fish of the desired ages became available, recruitment of various magnitude occurred in each population over a period of several weeks. This resulted in a heterogeneous age mixture of fry comprising each population.

Consequently, our examination of growth has to be restricted to general changes in size over time for all the populations. Fig. 10 illustrates such changes in length for the four populations. The populations were fed "ad libidum" six times/day, which amounted to a feeding rate of approximately 3% BW/day. Water temperatures for the rearing period are shown in Fig. 11. The combination of these data allows us to construct a crude standard by which we can begin to judge how much food and time will be required to raise chum salmon fry to various sizes under environmental conditions similar to those experienced by our populations.

The determination of optimum rearing conditions and the effects of size and timing of release on survival to the adult stage were not examined. Such questions are of obvious concern to ocean ranches and may have to be solved on an individual stock basis through the tagging or marking (e.g., electrophoretic markers) of arbitrary but discrete segments of a cultured population.

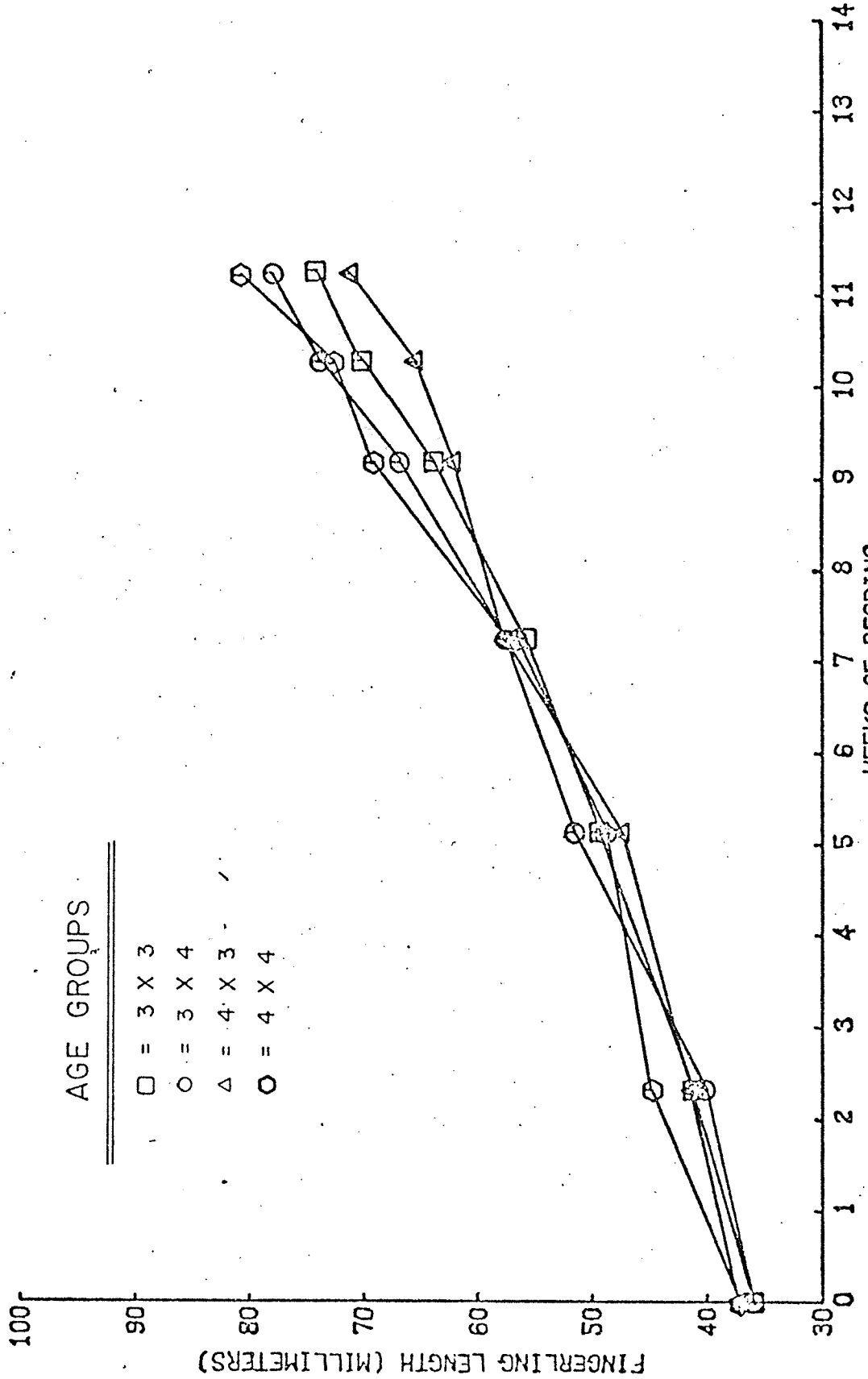


Fig. 10. The growth rates (changes in fork length) experienced by four different populations of chum salmon fry while reared in freshwater at Big Beef Creek.

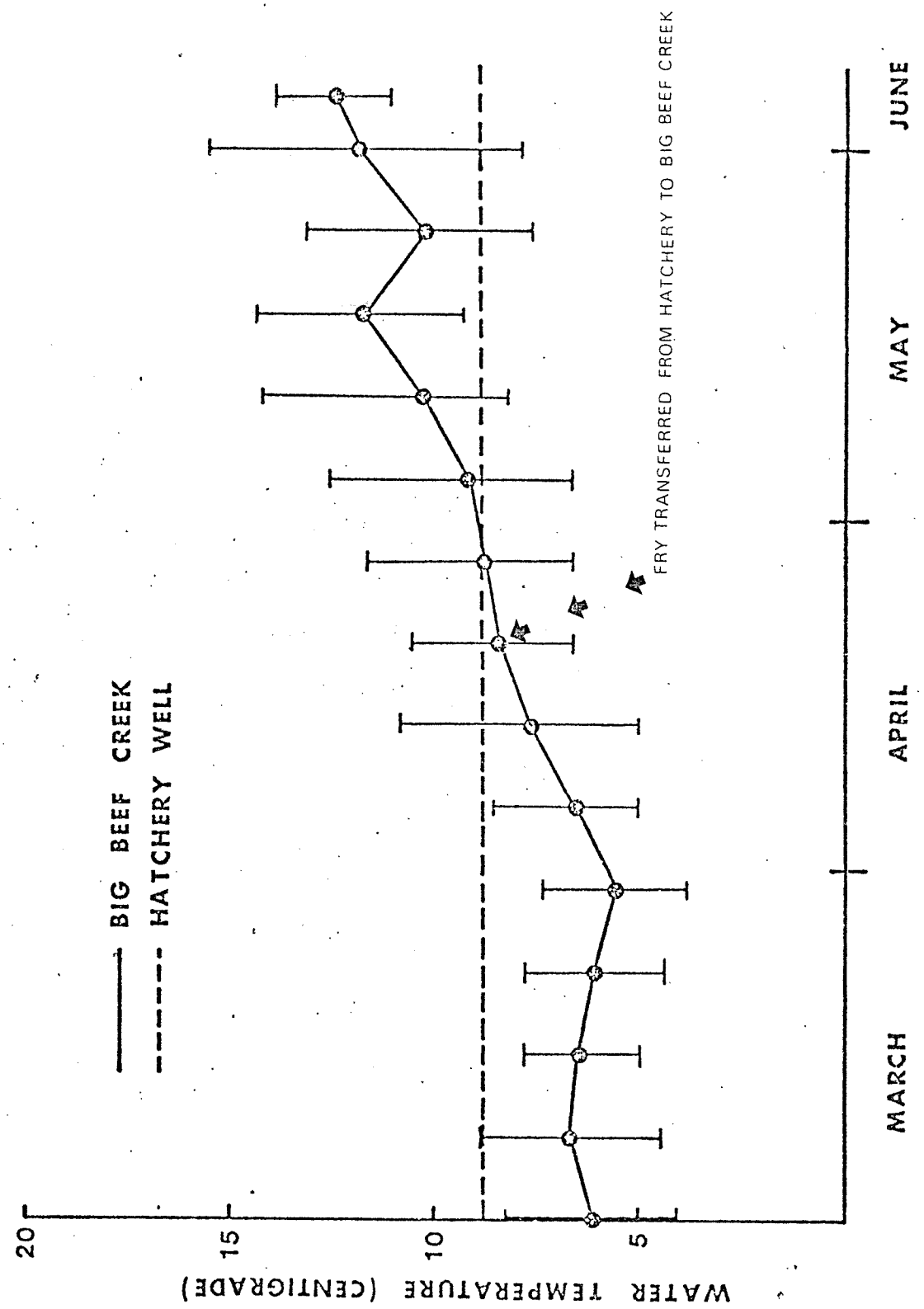


Fig. 11. Variations in the mean water temperature experienced by reared chum salmon populations while in the hatchery and at Big Beef Creek.

GENERAL CONCLUSIONS

Ultimately, the strategies employed in operating a chum salmon ocean ranch are going to be based upon the kind of fish that is desired. Because fry survival and quality can, to a certain extent, be controlled during the incubation phase, our attention should be directed toward the development of genetic strains which have a high potential to survive in the marine environment. Any deliberate manipulation of genes within our enhanced populations will be controlled by what we want from our cultured stocks and by what they are capable of giving us.

In developing such a program, the first step is the delineation of the genetic variability existing within the population and the determination of whether we can effectively utilize some of it to produce fish with desirable characteristics. Unfortunately, our comprehension of the impact of environmental factors on various genotypes is almost nonexistent. Feedback of this type is essential in the development of rational breeding schemes and will most likely be accumulated through trial and error. Questions relating to chum salmon culture, such as whether to have extended rearing, with feeding and under what conditions, remain to be answered.

During its evolution, the species may have been confronted with life-history options that may have been analogous to some to which we may expose them. It seems logical to expect that the life-history pattern of a stock is the most efficient given the environmental circumstances under which it has developed. Consequently, we are faced with trying to evaluate the impact of our culture techniques on the traditional life-history pattern. We must ask ourselves what impact will the removal of certain selection pressures make upon the potential fitness of the populations.

At present, it would seem prudent to perturb the natural system in small increments and evaluate the impacts as they occur. We have the time to do so. The potentials of careful appraisal far outweigh the possible catastrophic consequences of disarranging a sophisticated and delicate natural system.

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