AN ABSTRACT OF THE THESIS OF

DEREK CLINTON POON (Name) for the MASTER OF SCIENCE (Degree)

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Title: DEVELOPMENT OF A STREAMSIDE INCUBATOR FOR CULTURE OF PACIFIC SALMON

Abstract approved: Redacted for privacy
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Criteria for the design and construction of a prototype stream-side incubator for Pacific salmon (Oncorhynchus) eggs and alevins are described. Operation of the prototype and methods of transporting freshly spawned salmon eggs from distant sources are reported.

Effects of water velocity, stocking density, and substrate type on the growth, development, survival, and behavior of Pacific salmon embryos, alevins, and fry were studied. The results suggest that the optimum conditions result from low stocking density of about one layer of fertilized eggs, water velocity of 54 to 220 cm/hr, and a smooth cobble substrate.

The prototype incubator was tested with about 500,000 salmon eggs in 1968-69. Due to some serious mechanical problems and poor water circulation in the hatchery tanks, mortalities exceeded the fry production for the first year of operation. Several problems were

identified and design improvements suggested.

A method was developed for transporting live salmon eggs unfertilized by packing the eggs and milt in separate containers. By maintaining temperatures of 6°C and effecting fertilization after 20 hours in storage, very high (> 90%) fertility was obtained. A practical application of the method was demonstrated with 150,000 pink salmon eggs transported from southeastern Alaska to Oregon.

Development of a Streamside Incubator for Culture of Pacific Salmon

by

Derek Clinton Poon

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DEVELOPMENT OF A STREAMSIDE INCUBATOR FOR CULTURE OF PACIFIC SALMON

INTRODUCTION

Pacific salmon, genus Oncorhynchus, constitute one of the world's important seafood resources. In the Pacific Northwest, some salmon stocks have declined so alarmingly that artificial propagation has become a cogent agent in sustaining the native stocks.

In Oregon, vigorous hatchery programs supplement important coho (O. kisutch) and chinook (O. tschwatscha) salmon runs. Relatively little attention, however, has been given to hatchery propagation of pink (O. gorbuscha), chum (O. keta), and sockeye (O. nerka) salmon.

Pink salmon spawners have never been reported to be abundant in Oregon coastal streams, although they are commonly harvested in the local coastal commercial and recreational fisheries; presumably these fish are from Washington and British Columbia streams. Chum salmon, on the other hand, were once abundant in spawning streams on the north Oregon coast. Their numbers have become drastically reduced in recent years.

Unlike the other species of Pacific salmon which utilize fresh water as a nursery area, pink and chum salmon leave for the ocean as unfed fry soon after they emerge from spawning beds. Because of this characteristic, artificial propagation of these species

conceivably would require less time and cost than for the other salmon species which remain in fresh water as juveniles for several months.

The Japanese produce about 400 million chum salmon fry annually to support a fishery that harvests between 20 and 30 million pounds annually on Hokkaido Island (Kisaburo, 1965), more than a normal salmon harvest from the entire Oregon Coast. Although less than one percent of the hatchery fry survive to return as maturing adults, the Japanese consider their program to be highly successful; they allow only a small percentage of the escapement to spawn naturally.

In summer 1968, the Oregon Agricultural Experimental Station and the Fish Commission of Oregon initiated a program for the production of pink and chum salmon in a prototype hatchery, called a "streamside incubator." The hatchery is located on Whiskey Creek which flows into Netarts Bay, Oregon.

Although artificial propagation greatly increases fresh water survival, hatchery fry may exhibit a pronounced inferiority to wild fry in survival potential (Foerester, 1938). This difference might be explained by the artificial conditions to which salmon are subjected in the hatchery. One objective of this study is to produce salmon fry showing good growth and possessing behavioral traits similar to wild fry.

Under natural conditions, eggs and alevins repose under a protective layer of gravel, and water flow is laminar. Early development occurs in darkness, and there is little physical activity until salmon begin to emerge as free-swimming fry.

In typical hatchery operations, eggs are concentrated in trays which are placed in horizontal troughs or stacked vertically. The water flow is turbulent as it passes through the trays; re-aeration is usually provided between trays. Alevins may be held in incubation trays until after their yolk has been absorbed, or they may be transferred to open troughs or ponds to complete their development. The fish are often exposed to artificial or natural light prematurely.

The obvious differences between the hatchery environment and the natural environment may produce observable differences in fry quality. Brannon (1965) reported that the high water velocity through a hatchery incubator forces the alevins to exercise prematurely at the expense of growth. He also found that exposure to light retards development, causes increased mortality, and slows yolk absorption. In comparing various artificial incubation techniques with natural incubation, Bams (1967) found that the largest fry resulted from natural incubation and from incubation schemes which simulated natural conditions. According to Bams, the standard hatchery techniques not only produced a smaller fry, but one which showed a lessened capability to perform in swimming and predation tests also.

Preliminary experiments were run at the Marine Science Center, Newport, in 1967-68 to establish design criteria for substrate type, water velocity, and stocking density in a streamside incubator (McNeil, 1968). The prototype was built at Netarts Bay in 1968 and was designed to handle one million eggs reposing on a crushed rock substrate. Water velocity past the eggs was set at about 25 cm/hr. Further experiments were run at Netarts Bay in 1968-69 to refine the design criteria established by the preliminary experiments in 1967-68.

A second objective of this study was to determine if non-native stocks of pink and perhaps chum salmon could be introduced successfully to Oregon. We thus require the capability to transport salmon eggs to Oregon from locations where surplus fish are available. Because Alaskan streams are likely sources of eggs, it was decided to explore the possibility of acquiring eggs at remote locations and transporting them directly to Oregon.

One traditional method of handling freshly spawned eggs is to fertilize them at the collection site, allow the eggs to water harden for a few hours, and ship them in iced trays before they become oversensitive to shock. Another method, not in common use, is to store male and female gametes in separate containers during shipment and effect fertilization at the destination.

Methods of delayed fertilization were studied in summer of 1968 as a part of this thesis research. Pink salmon eggs were transplanted

from Southeastern Alaska to Oregon in September 1968, to demonstrate the practical application of delayed fertilization. To further substantiate the feasibility of using the delayed fertilization method, two subsequent field tests were run in October and November 1968 with coho salmon, replicating the techniques used in the transportation of pink salmon gametes. Surplus coho eggs were provided by the Fish Commission of Oregon for these tests.

This thesis first reports the results of experiments intended to establish criteria for the design and operation of a streamside incubator. It next describes the construction and operation of the prototype incubator. Finally, the thesis reports on methods for transporting freshly spawned eggs over long distances.

This research was supported in part by the Fish Commission of Oregon, the U.S. Bureau of Commercial Fisheries (Alaska Region), and the National Science Foundation's Sea Grant Program.

CRITERIA FOR THE DESIGN AND OPERATION OF A STREAMSIDE INCUBATOR

Under natural conditions, salmon eggs and alevins repose within a gravel substrate where water flow is laminar (Vaux, 1967). To survive, eggs and alevins must remain undisturbed, and be free of predators, parasites, and pathogens. The water flow must supply sufficient oxygen to the chorion of each individual egg to satisfy its metabolic requirements and must remove the toxic wastes of metabolism from its immediate vicinity; sufficient intragravel flow therefore is imperative for proper growth and development of embryos and alevins.

The design of an incubation system must also take into account stocking density. Eggs and alevins are not only in competition for the available dissolved oxygen, but they also release waste metabolites which become toxic under certain conditions. Overcrowding in hatchery tanks could well impair growth and development and create conditions favorable for disease organisms and fungus.

The velocity of intragravel flow can be approximated by using field measurements of permeability (k) and hydraulic gradient (i) in the equation:

$$V = ki \qquad (1)$$

Equation (1) gives apparent (superficial) velocity (V) which can also be defined as the discharge (Q) per cross sectional area of streambed

(A), i.e.

$$V = ki = \frac{Q}{A} \qquad (2)$$

Since the spawning bed is made up of more than 80 percent of solid materials, and since water velocity increases when streaming around particles, the average true velocity is at least five times the calculated apparent velocity.

Field measurements suggest that streambed permeability may vary between 2,400 and 18,000 cm/hr (Wickett, 1958; McNeil and Ahnell, 1964). The hydraulic gradient of typical pink and chum spawning beds ranges from 0.1 to 0.7 percent (McNeil, 1966; McNeil, personal communication). Using the above measurements of k and i in equation (1), the calculated apparent velocity through a natural spawning bed ranges from 2.4 cm/hr to 126 cm/hr. In a pink salmon spawning bed with a gradient of 0.3 percent, Vaux (1967) found the apparent velocity to be 32 cm/hr by direct measurement.

A commonly recognized factor limiting survival in natural spawning beds is oxygen privation caused by insufficient water seepage in the gravel. High mortality of salmonid embryos has been observed in natural redds where oxygen level and intragravel flow were low (Wickett, 1954). High mortality also resulted when embryos were placed in porous containers and buried in gravel beds where oxygen level and water velocity were low (Phillips and Campbell, 1962). In laboratory experiments, reduced water velocity produced

newly hatched steelhead trout (Salmo gairdneri) and chinook salmon which were smaller and weaker than control fish; these effects were most pronounced at the lower dissolved oxygen levels tested (Silver, Warren and Doudoroff, 1963). The work of Shumway, Warren and Doudoroff (1964) with steelhead trout and coho salmon produced similar results.

Design criteria for an incubation system must take into account the minimum water velocity which will satisfy the dissolved oxygen requirements of eggs and alevins, yet not exceed a velocity beyond which the alevins may be forced to exercise excessively. Generally, the supply of dissolved oxygen to an embryo is both a function of the dissolved oxygen content and the water velocity. Observations on Atlantic salmon (S. salar) by Hayes, Wilmot and Livingston (1951) and on chum salmon by Alderdice, Wickett and Brett (1958) suggest that the rate of oxygen consumption per unit mass of embryonic tissue is little affected by growth over most of the period between fertilization and hatching. Therefore, the rate of oxygen consumption by a population of embryos could be a simple function of metabolic rate which increases as development progresses. Consequently, it may be advantageous to increase water velocity as the time for hatching approaches.

On the other hand, the turbulent water flow, such as occurs under standard hatchery conditions, may be harmful by forcing alevins to

exercise prematurely. Although hatchery methods yield high survival, the resulting fry may be smaller than wild fry and show a lessened capacity to perform work (Bams, 1967).

In nature, the gravel substrate protects the eggs and alevins from predation, light, and dislodgment. Effects of substrate on growth, development, and behavior have not been well defined, however.

When brown trout (S. trutta) were reared on a substrate of different rugosity, Marr (1963, 1965) found that alevins reared on a grooved surface were heavier, converted yolk into body tissue more efficiently, and were less active than alevins reared on a smooth surface; alevins on the smooth surface were more active in their attempts to maintain an upright posture. Because traditional methods of rearing salmon on screen trays may not satisfy the tactile requirements of developing alevins, the type of a substrate may be an important consideration in the design of an incubation system. Hence, the type of substrate was studied along with water velocity and stocking density to design a hatchery system which would produce a robust fry retaining behavioral traits typical of wild fry.

Effects of water velocity, egg density, and substrate characteristics on growth and survival of eggs and alevins were studied by conducting a series of experiments which were initiated in autumn 1967 and terminated in spring 1969. Preliminary experiments at the

Marine Science Center, Newport, in autumn and winter 1967-68 established initial criteria for design of the prototype incubator constructed at Netarts Bay in summer 1968. Further experiments at Netarts Bay in autumn and winter 1968-69 provided a basis for refining the original criteria.

Experiments at the Marine Science Center, Newport, Oregon (1967-68) (McNeil, 1968)

To determine more precisely the water velocity requirements of eggs and alevins, test lots of newly fertilized chum salmon eggs were held in aerated water flowing at velocities of 6.5, 12.5, 25, 50, and 100 cm/hr. Ten experimental cells were immersed in a water bath, and the amount of water passing through each cell was regulated by adjusting the difference in head (Δh) between the water bath and the outflow from a drain at the base of a cell (Figure 1).

Each cell had a cross-sectional area of 433 cm^2. The substrate was crushed gravel 15 cm deep. Eggs placed on the surface of the substrate were from a mixed lot originating from eight chum salmon females.

The egg-bearing females were collected from Whiskey Creek on November 15, 1967. Six females were partly spawned when collected and two were unspawned. Sperm was collected from several partly spawned males and placed in a container separate from the eggs. Eggs and sperm were transported to the Marine Science Center, 70

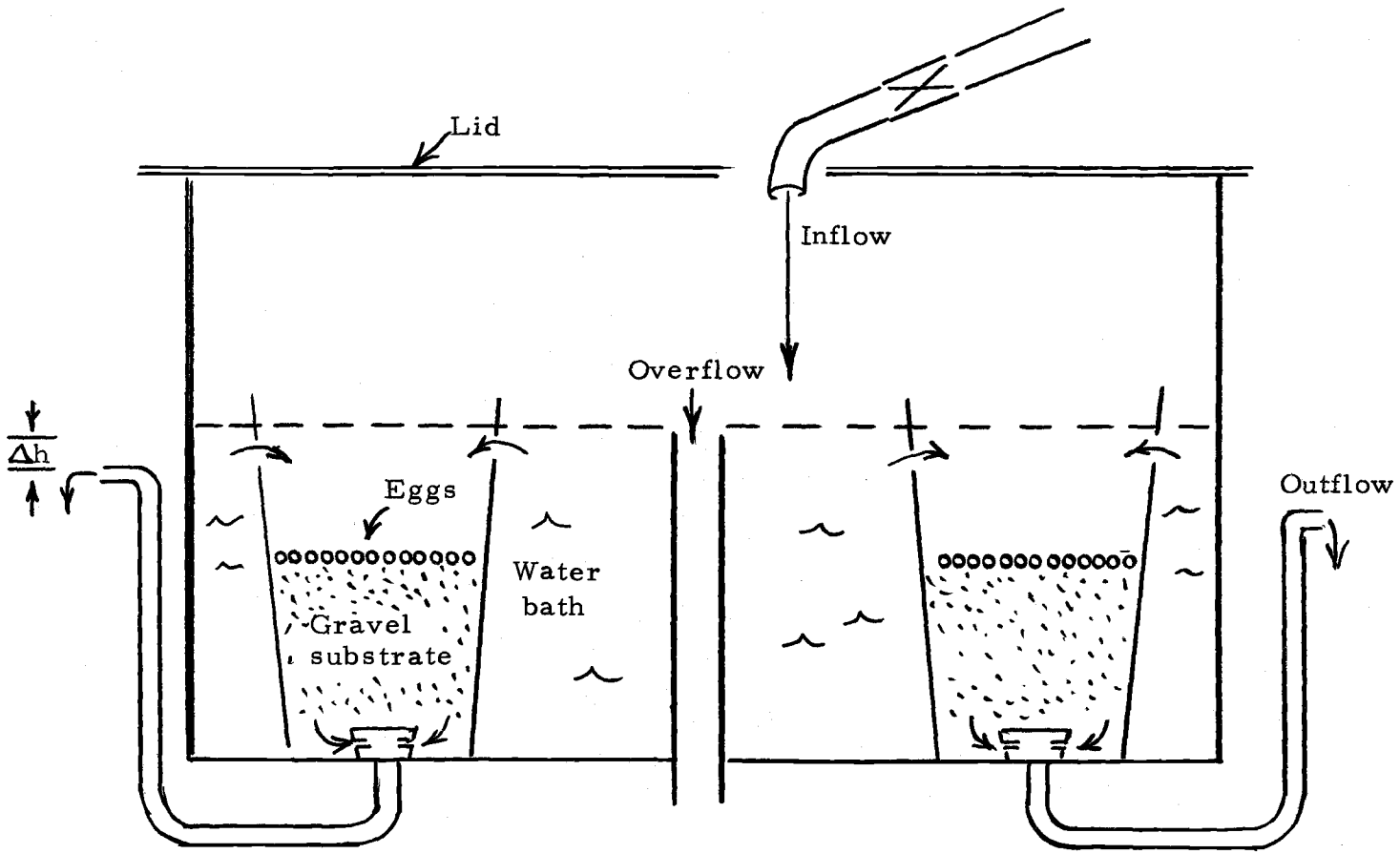

Figure 1. Schematic diagram of water bath and experimental cells.

miles south of Netarts Bay, and fertilization was accomplished three hours after the time of collection. About 1200 randomly chosen eggs were placed in each of the ten cells.

Flow rates were adjusted at 30 cm/hr in each cell for the initial 72-hour period after fertilization. It was apparent at the end of 72 hours that a high percentage of the eggs were unfertilized. Of the original 1200 eggs in each cell, the number unfertilized ranged from 42 to 49 percent of the total. Thus, the number of fertilized eggs in each cell ranged from 600 to 700 initially. The unfertilized eggs were believed to have originated mostly from the two unspawned females, because relatively few of the eggs from these fish were loose in the body cavity at the time of collection.

Five test cells were grouped on either side of the water bath, and the five test velocities were replicated in each group of five cells. The average of 75 velocity measurements for each cell indicated that the desired test velocities were closely approximated over the incubation period (Table 1).

The fertilized eggs hatched over the period January 14-16, 1968. The alevins in cells numbered 1 through 5 were covered with crushed gravel on January 16; whereas, those in cells numbered 6 through 10 were allowed to remain on the surface of the substrate. Water velocity remained unchanged. Purpose of adding the gravel to one test group was to compare the effect on growth and development of

Table 1. Average velocity (in cm/hr) of water flow in test cells over the period of incubation up to hatching.

Cell number	Velocity (average of 75 observations)
1	6.5
2	12.6
3	23.6
4	49.7
5	100.5
6	6.6
7	13.3
8	25.5
9	50.2
10	97.8

confinement within substrate materials, with no confinement.

There was no significant difference in the percentage of eggs hatching or the time of hatching over the range of water velocity tested (6.5 to 100 cm/hr). Size of alevins was, however, significantly affected by water velocity.

The diameter of the eye is used to demonstrate differences in the size of alevins. Unpublished studies with pink salmon embryos and alevins show that size of the eye correlates well with various other anatomical measurements, including size of body, pectoral fins, and mid brain (Ralph Wells, personal communication).

There was a highly significant effect of water velocity on eye diameter at hatching ($F = 3.66$ with 4 and 95 degrees of freedom). Newly hatched alevins reared at water velocity of 6.5 cm/hr had a

significantly smaller eye than those reared at higher velocities. There was no significant difference in eye diameter at 25 cm/hr and above, however.

Measurements of eye diameter 20 and 65 days after hatching indicated that the initial small eye diameter of alevins reared at 6.5 cm/hr persisted to absorption of the yolk. Figure 2 shows the influence of water velocity on the mean eye diameter from hatching to 65 days after hatching.

There was also some suggestion that alevins reared at 50 cm/hr were longer and heavier than those reared at lower and higher velocities. Mean body length of 10 alevins collected from test cells 65 days after hatching are plotted in Figure 3. There was no apparent difference in weight or length between alevins buried in the gravel (Group #1) and alevins on the surface of the gravel (Group #2).

A second experiment was conducted in late winter 1958 with the apparatus at the Marine Science Center to gain some insight into the effects of crowding on survival, growth, and development. Steelhead eggs from the Oregon Game Commission Alsea Hatchery were used in experiments on crowding. Eggs were taken by hatchery personnel from 11 unspawned females on March 5, 1968, and transported about 50 miles to the Marine Science Center where they were fertilized with milt from two males and mixed. Flows in nine test cells (433 cm^2 surface area) were adjusted to 25 cm/hr, and the following numbers

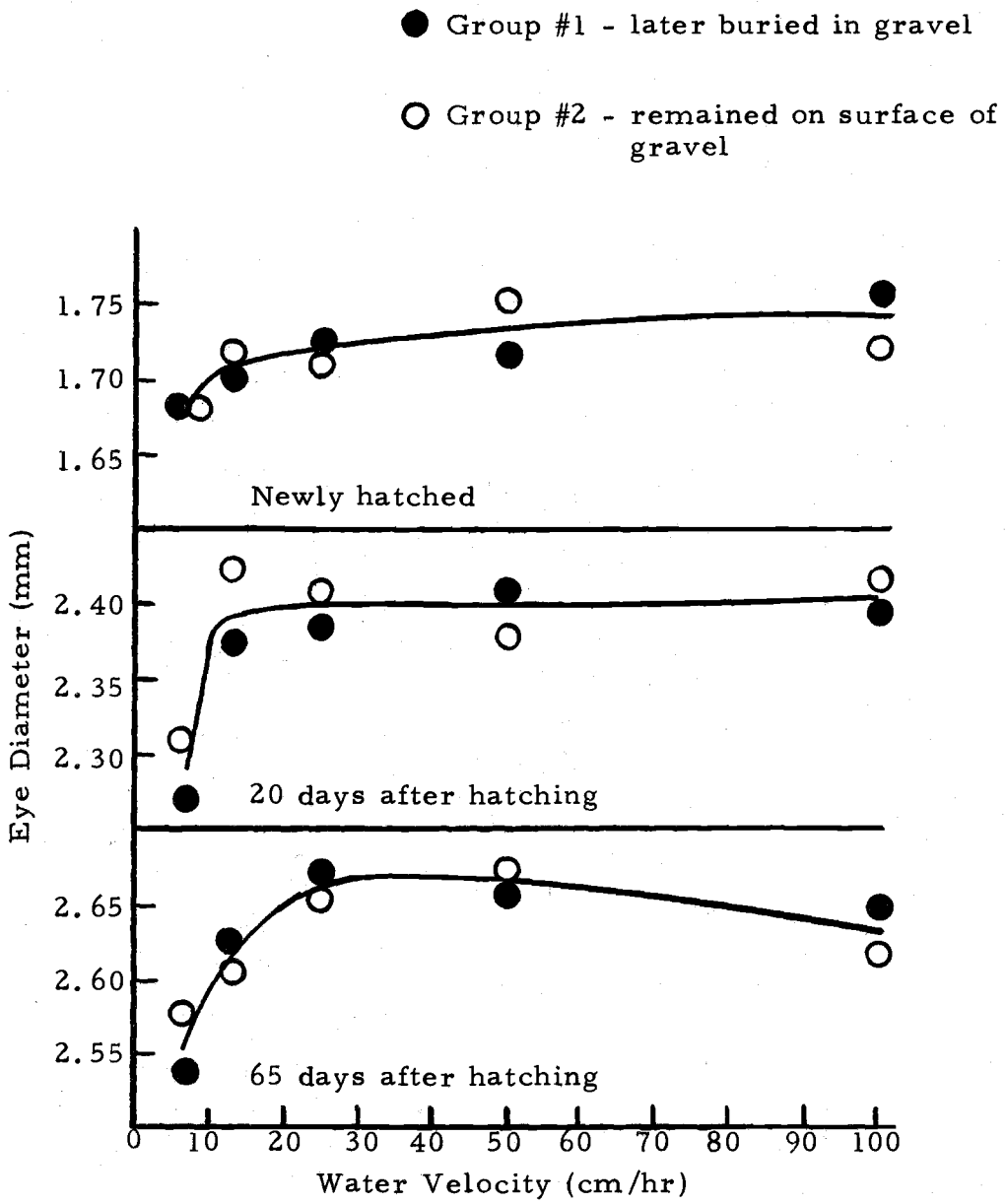

Figure 2. Influence of water velocity on the mean eye diameter of chum salmon from hatching to 65 days thereafter.

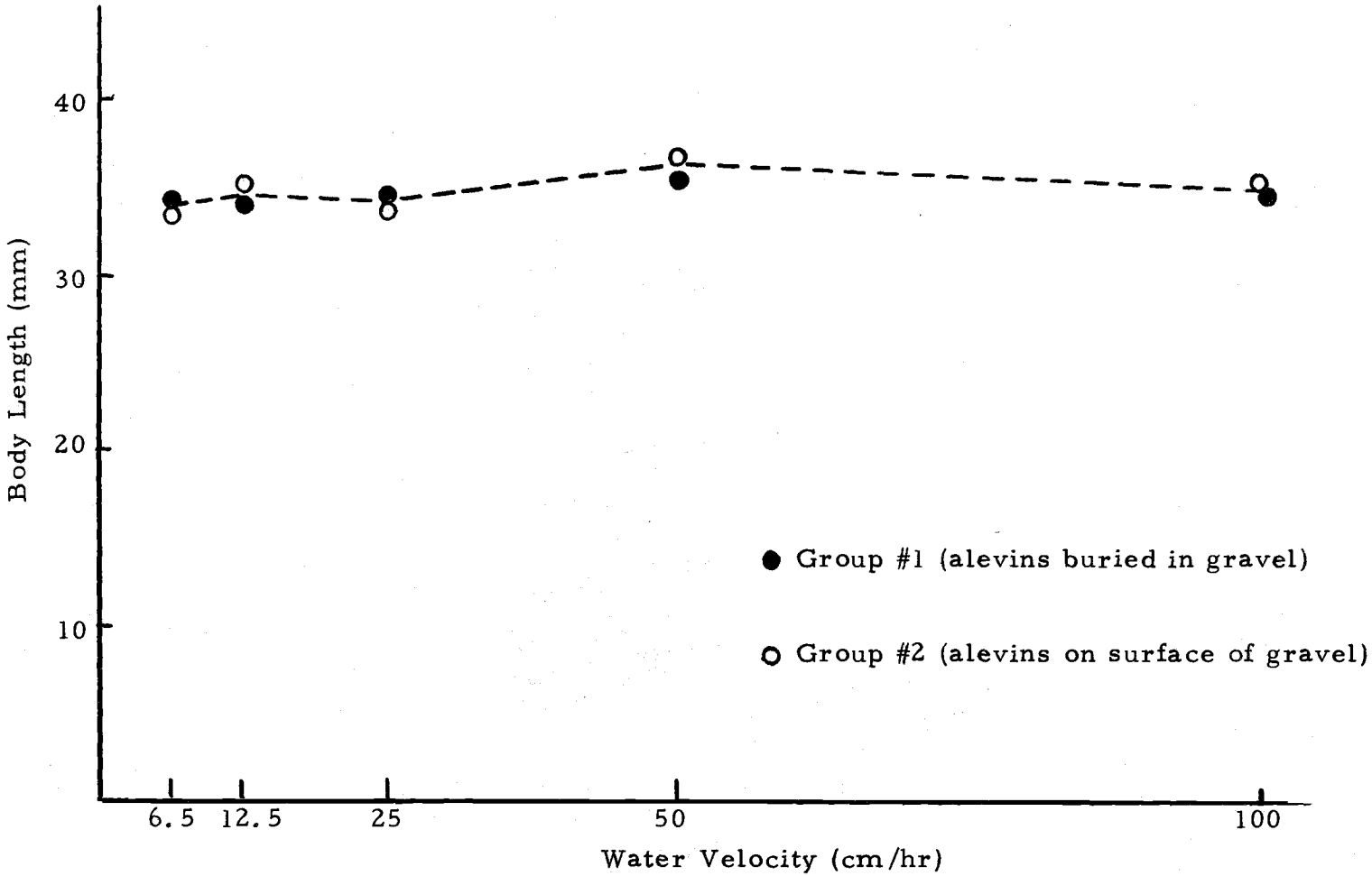

Figure 3. Mean body length (tip of snout to fork of tail) of alevins 65 days after hatching.

of eggs were placed in the cells:

Cell #1 - 500 eggs	Cell #6 - 5,000 eggs
#2 - 1,000	#7 - 6,000
#3 - 2,000	#8 - Not used
#4 - 3,000	#9 - 7,000
#5 - 4,000	#10 - 8,000

The eggs hatched on about April 5 with less than five percent mortality in any of the test cells. However, after hatching there was heavy mortality (50 percent or higher) in all cells with 4,000 or more eggs. The mortality was first observed in the cell with 8,000 eggs (April 11) and last observed in the cell with 4,000 eggs (April 25). Less than 20 percent mortality occurred in the cells with fewer than 4,000 eggs:

No. eggs in cell	Mortality to yolk absorption
500	17%
1,000	5
2,000	8
3,000	7

Mean net weight of the surviving fry was determined for the five cells with initial egg densities of 4,000 per cell and less. The measurements were made on May 7, May 14, and May 28. The fry were fed over the period May 8-28 and exhibited considerable growth. The results are summarized in Table 2.

The experiment clearly demonstrated that densities of 2,000 steelhead eggs per cell (about five per cm^2) and higher cause retarded growth at a water velocity of 25 cm/hr. Even though fry were fed excess quantities of food over the three-weeks of May 8-29, the same

Table 2. Mean wet weight of steelhead fry cultured in test cells with water velocity held at 25 cm/hr.

Initial density of eggs in cell	Wet weight on date		
	May 7 (mean of 20 fry)	May 14 (mean of 10 fry)	May 28 (mean of 10 fry)
500	220 mg	260 mg	330 mg
1,000	217	260	330
2,000	211	242	283
3,000	188	210	267
4,000	164	205	273

relative difference in size of fry persisted; the smaller fish did not gain on the larger fish.

Absorption of yolk was also delayed in cells where the eggs and alevins were most concentrated. On May 6, only 20 percent of the fry in cells nos. 1 and 2 (500 and 1,000 eggs) exhibited external yolk. This compared with 25 percent in cell #3 (2,000 eggs), 50 percent in cell #4 (3,000 eggs) and 75 percent in cell #5 (4,000 eggs).

Results of the preliminary experiments at the Marine Science Center provided criteria on water velocity and density of eggs and alevins for a prototype streamside incubator. Water velocity in the range of 13 to 100 cm/hr appeared to be suitable for high survival, provided excessive crowding of eggs was avoided. The preliminary tests suggested that 25 to 50 cm/hr may possibly afford conditions for maximum growth and survival. The density of eggs of the size of steelhead probably should not exceed five per cm^2 (about 4,500 per

ft^2) at a water velocity of 25 cm/hr.

Although these early experiments did not consider substrate characteristics, it appeared that crushed rock was suitable as a substrate. Furthermore, there was no evident need to bury the eggs and alevins in the substrate. This greatly simplified the design and operation of the incubator.

Experiments at Netarts Bay, Oregon (1968-69)

The experiments at Netarts Bay in 1968-69 refined the design criteria established from the experiments at the Marine Science Center during 1967-68. The effects of water velocity, stocking density, and substrate type on growth and behavior of Pacific salmon were investigated further; but in refining the original criteria, the Netarts study provided (1) a wider range of water velocity, (2) different substrates to gain insight into the tactile requirements of developing alevins, and (3) simultaneous observations on the three factors (velocity, substrate, and crowding) to allow for evaluation of possible interactions.

Each factor was tested at three levels. Water velocity was set at 13, 54, and 220 cm/hr; eggs were stocked at 1.5, 2.4, and 3.3 per cm^2; substrates were nylon screen (supported by perforated plywood), crushed rock (1/4 to 1/2 inch diameter), and smooth cobble (1 to 2 inch diameter). The factorial design gave 27 possible

combinations to be tested (3 x 3 x 3).

Twenty-seven plastic experimental cells were installed in two hatchery tanks (Figures 5 and 6) at Netarts Bay. Each cell (Figure 4) had a cross-sectional area of 725 cm^2. Water velocity was controlled by adjusting the head difference between the water bath and the outflow tube, similar to the experimental cells used at the Marine Science Center the previous year. Experimental cells were stocked with coho, chinook, and chum salmon eggs.

A second experiment was run to determine if eggs incubated in standard Heath incubators before being transferred to a natural substrate produced fry similar in quality to fry resulting from eggs reared on a natural substrate from the date of fertilization. Several thousand coho eggs were incubated in the Heath incubator until the eyed stage; about 700 eggs were then placed into each of three experimental cells prepared with the three test substrates. Water velocity was set at 54 cm/hr.

To remove silt from the hatchery water supply, water was passed through a series of wooden baffles with holes drilled alternately on top and on the bottom (Figure 7). The filter directs the water downward and upward through two layers of oyster shells and two layers of crushed rock before entering the experimental cells. The filter was partially effective for two months, but it was bypassed thereafter because of clogging.

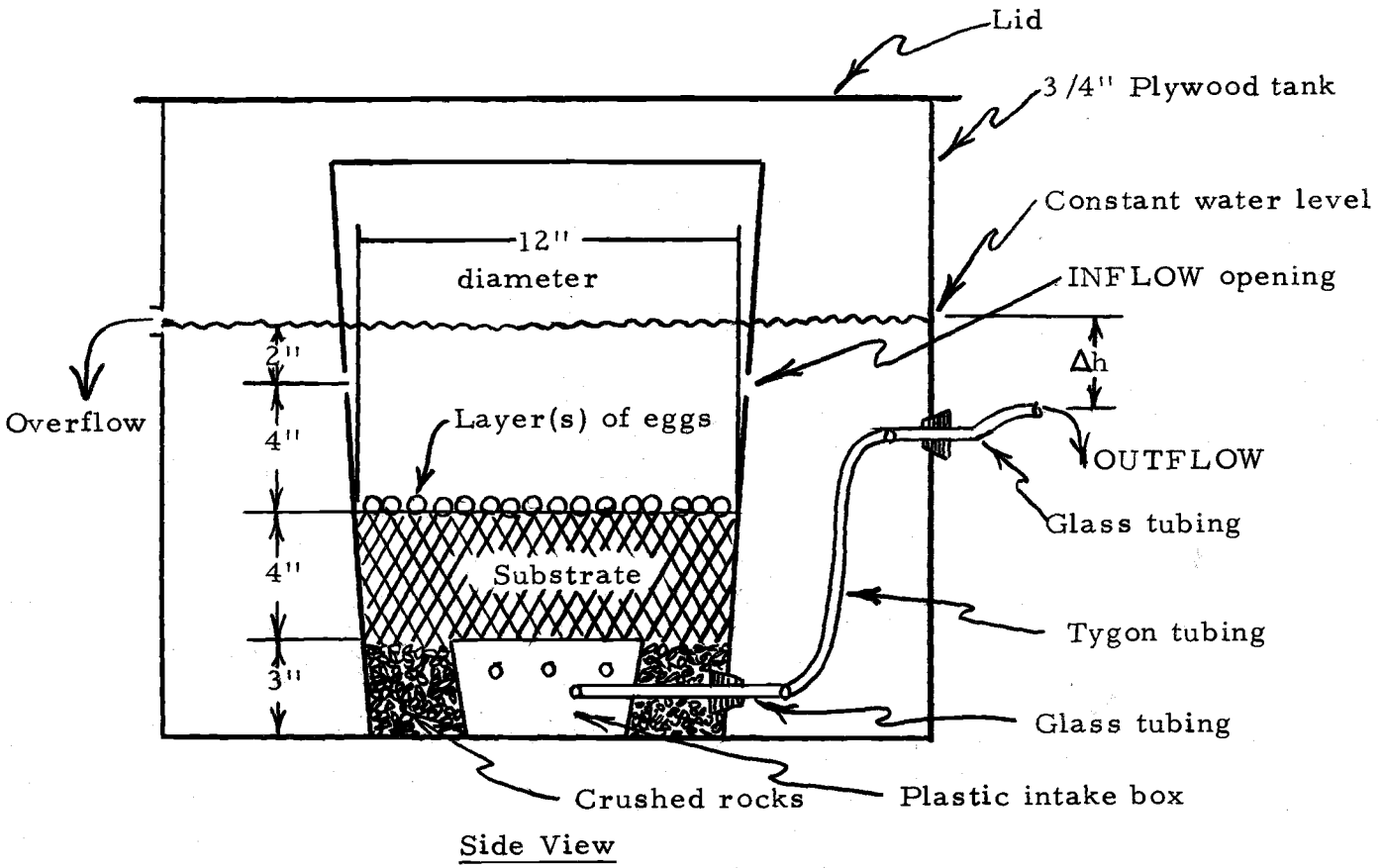

Figure 4. Schematic diagram of an experimental cell showing direction of water flow.

Figure 5. Experimental cells and filter tank.

Figure 6. Closeup of experimental cells.

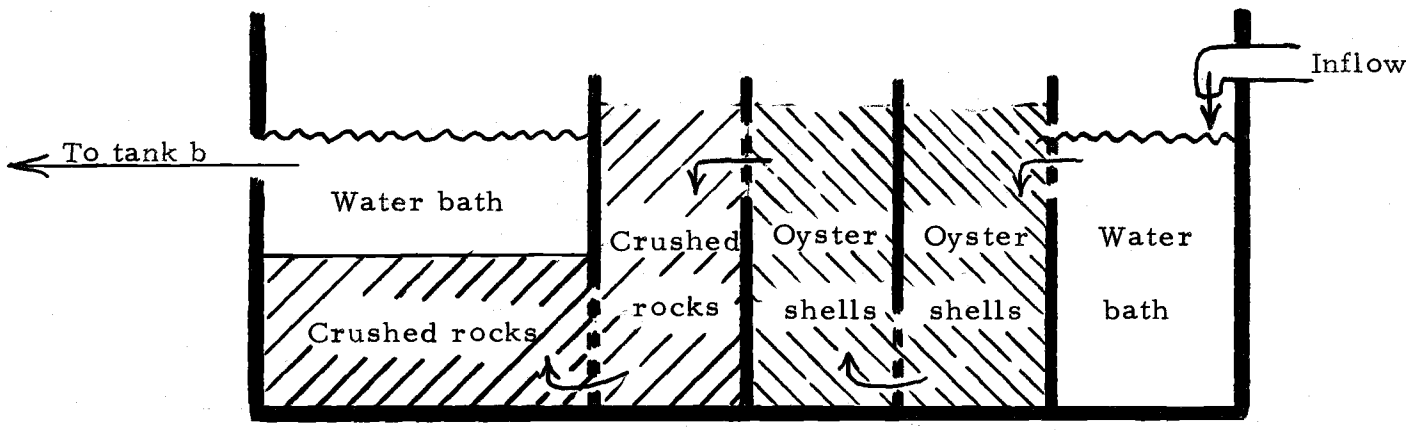

Figure 7. Schematic side view of filter tank (a) which precedes hatchery tanks (b and c) housing the experimental cells.

All water velocities were recorded and reset where necessary at least every third day. Eggs were treated with a fungicide (malachite green) twice weekly until hatching to reduce fungus growth on dead eggs.

Experimental cells were stocked on November 25-27 with 700, 1,400 and 2,000 newly fertilized coho eggs from the Oregon Fish Commission Trask River Hatchery. Equal numbers of eyed eggs were later introduced into each cell; 200 chinook eggs acquired from Dr. Lauren Donaldson at the University of Washington were introduced on December 17 and 200 chum eggs acquired from Whiskey Creek were introduced on January 4. An estimated 1,400 coho and 1,700 chinook eggs were held in a Heath incubator for further comparisons.

After the first eggs were introduced to the experimental cells, all velocities were held at about 50 cm/hr for 72 hours before being adjusted to the prescribed test velocities.

Growth and development was evaluated from measurements of eye diameter and total length of eyed embryos, alevins, and "swim-up" fry. Eye diameter proved to be the more sensitive measurement and was used as the criterion for growth in all statistical analyses. A strong correlation between eye diameter and total length was found in tests with coho salmon fry ($r = 0.841$) (Figure 8).

Analysis of variance was used to test for significant differences in eye diameter among the various treatments. Values of least

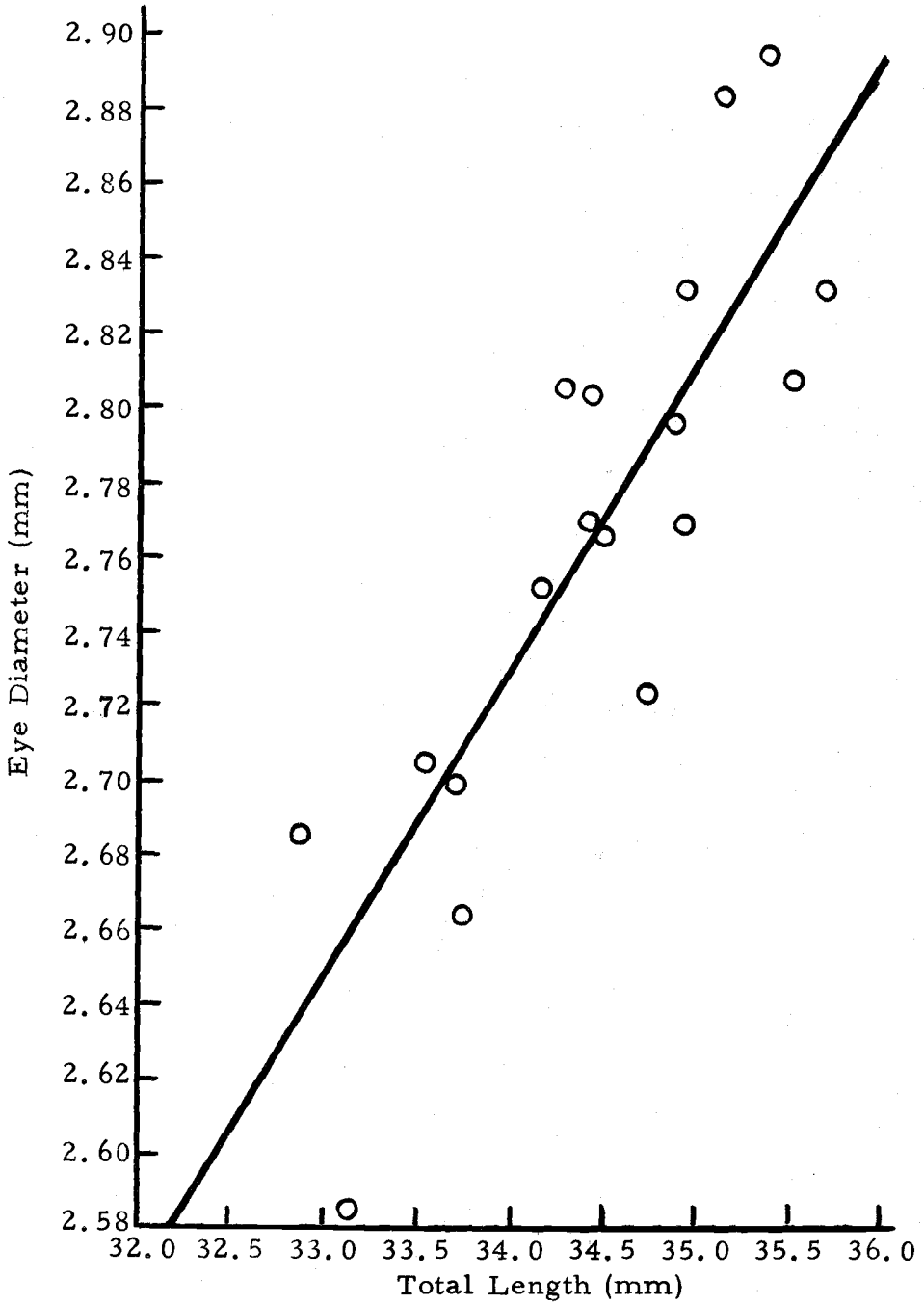

Figure 8. Linear regression between eye diameter and total length of coho fry. The calculated regression equation is $Y_i = .06 + (.082)X_i$.

significant difference (LSD) were calculated to separate the treatments which produced the larger fish from those which produced the smaller fish. Conclusions were based both on the results of the more formal statistical tests as well as empirical examination of trends and patterns revealed by the results of the experiments.

Problems with the water delivery system and with the water circulation in the experimental cells made it difficult to maintain some of the planned experimental conditions. During freshets, water pumped from Whiskey Creek contained fine silt which became deposited on eggs and clogged interstices in the substrate. Coarse sand debris occasionally collected in the impeller of the water pump and impaired its performance. Infrequent power failures caused water to stop for periods of up to two hours. Muddy water made dead eggs difficult to discern, and when heavy mortalities occurred in some experimental cells, their removal was time consuming. Treatments with malachite green failed to erradicate fungus which further impaired water circulation. Test cells were periodically backflushed with air, and it was common practice to set velocities about 10 percent higher than scheduled to accommodate an expected drop in velocity between readings.

Although the water velocity fluctuated greatly at times, all cells were affected equally. Based on the averages of 83 velocity readings (Table 3) throughout the experiment, the desired velocities were closely approached.

Table 3. Average water velocities in experimental cells with various stocking densities and substrate types.

Stocking Density	Substrate Type	Average Water Velocity[1/]
eggs/cm^2		cm/hr
1.5	screen	200.1
1.5	crushed rocks	202.0
1.5	smooth cobbles	202.9
1.5	screen	53.0
1.5	crushed rocks	53.3
1.5	smooth cobbles	53.2
1.5	screen	13.0
1.5	crushed rocks	13.2
1.5	smooth cobbles	13.3
2.4	screen	200.3
2.4	crushed rocks	204.8
2.4	smooth cobbles	196.0
2.4	screen	53.1
2.4	crushed rocks	52.8
2.4	smooth cobbles	53.2
2.4	screen	13.2
2.4	crushed rocks	13.0
2.4	smooth cobbles	12.7
3.3	screen	197.9
3.3	crushed rocks	197.2
3.3	smooth cobbles	194.1
3.3	screen	54.2
3.3	crushed rocks	52.8
3.3	smooth cobbles	54.1
3.3	screen	12.9
3.3	crushed rocks	13.5
3.3	smooth cobbles	13.1
	Supplementary Experiment with Eyed Eggs:	
1.5	screen	54.2*
1.5	crushed rocks	54.7*
1.5	smooth cobbles	53.0*

[1/] An average of 83 readings

* An average of 42 readings

Downward migration of alevins made the collection of some samples difficult, and the data were omitted. After hatching, alevins penetrated into the coarse cobble substrate, and some even penetrated beneath the nylon screens overlying the perforated plywood. Alevins penetrating into the coarse cobbles emerged later, but those penetrating beneath the screen were trapped and died. On several occasions it became necessary to remove decomposed alevins which clogged the screen and impaired the water flow. Consequently, sampling was not from the entire population in cells with the screen and smooth cobble substrates. Data on growth were collected for all cells, but data from cells where samples were suspect were not included in the analysis.

Evaluation of the effects of crowding and water velocity on eyed eggs and alevins was limited to observations from the cells with crushed rock, where little downward movement of alevins into substrate materials occurred. Data from cells with crushed rock and smooth cobbles were used for emerged fry. The planned comparisons were modified because of the failure to make observations from the screen substrate and from the smooth cobble substrate before fry emergence. A valid comparison was possible, however, with the post-emergence fry sample where crushed rock was compared with cobbles.

With coho and chinook salmon, no significant differences in hatching time were observed among the experimental cells. Size of

fish (eye diameter) and survival was influenced significantly by water velocity and stocking density but not by substrate type, which did, however, produce an observable difference in fry behavior.

Coho Salmon in Test Cells

Survival of coho salmon (stocked at time of egg fertilization) was lower than survival of chinook salmon (stocked as eyed eggs). Samples from all cells indicated that initial fertility was high. However, mortalities in excess of 50 percent occurred just before hatching in all cells stocked at medium and high densities; cells stocked at low density experienced less than 20 percent mortality. Mortality after hatching remained low in all cells.

Growth appeared to be influenced more by water velocity than stocking density. The best growth was exhibited by fry reared under low density and high water velocity of 200 cm/hr (see Tables 4, 5, 6, and 7). Conditions which produced favorable growth formed a clear convergent pattern (see Figure 9) over time beginning at the eyed stage when only the cells with 13 cm/hr and medium and high stocking density produced significantly smaller fish (i.e. eye diameter). Cells with high water velocity (220 cm/hr) and low stocking density produced the largest fry. The effects of water velocity on eye diameter of coho salmon during development is best illustrated by the low density cells with crushed rock substrate (Figures 10 and 11).

Table 4A. Eye diameter[1/] (in millimeters) of eyed coho salmon eggs reared at various water velocities and stocking densities and with different substrate.[2/]

Stage of Development	Stocking Density eggs/cm^2	Water Velocity	13 cm/hr		54 cm/hr		220 cm/hr
EYED EGGS	1.5	S	1.910±.038	S	1.899±.034	S	1.936±.023
	1.5	R	1.899±.040	R	1.959±.031	R	1.905±.034
	1.5	C	1.912±.054	C	1.876±.041	C	1.961±.036
	2.4	S	1.841±.038	S	1.941±.035	S	1.892±.053
	2.4	R	1.822±.050	R	1.992±.015	R	1.982±.023
	2.4	C	1.775±.050	C	1.943±.033	C	1.966±.026
	3.3	S	1.796±.035	S	1.859±.035	S	1.945±.037
	3.3	R	1.818±.042	R	1.961±.021	R	1.978±.023
	3.3	C	1.827±.039	C	1.905±.028	C	1.931±.037

[1/] Each is a mean of 20 measurements and is given with its 90% confidence interval.

[2/] Substrate types are represented by: S (screen), R (crushed rocks), and C (smooth cobbles).

Table 4B. Eye diameter and total length[1/] (both in millimeters) of coho salmon alevins and fry reared at various water velocities and stocking densities and with different substrates.[2/]

Stage of Development	Stocking Density	Water Velocity											
		13 cm/hr				54 cm/hr				220 cm/hr			
	eggs/cm^2	Eye Diameter		Total Length		Eye Diameter		Total Length		Eye Diameter		Total Length	
ALEVINS	1.5	R	2.293±.038	R	26.70±.45	R	2.478±.027	R	28.40±.29	R	2.501±.029	R	28.70±.36
	2.4	R	2.302±.041	R	26.95±.54	R	2.427±.046	R	27.95±.63	R	2.432±.044	R	28.20±.53
	3.3	R	2.237±.043	R	26.30±.38	R	2.387±.037	R	27.70±.49	R	2.482±.031	R	28.25±.43
	1.5	R	2.751±.039	R	34.20±.68	R	2.832±.022	R	34.95±.32	R	2.895±.021	R	35.40±.32
	1.5	C	2.700±.049	C	33.70±.80	C	2.804±.025	C	34.45±.51	C	2.884±.029	C	35.15±.68
FRY[3/]	2.4	R	2.585±.054	R	33.15±.81	R	2.809±.032	R	35.55±.51	R	2.770±.045	R	34.45±.82
	2.4	C	2.664±.024	C	33.75±.36	C	2.769±.028	C	34.95±.65	C	2.797±.035	C	34.90±.56
	3.3	R	2.686±.028	R	32.90±.38	R	2.723±.049	R	34.95±.75	R	2.805±.035	R	34.30±.53
	3.3	C	2.706±.030	C	33.55±.39	C	2.767±.027	C	34.50±.55	C	2.832±.041	C	35.70±.75

[1/] Each is a mean of 20 measurements and is given with its 90% confidence interval.

[2/] Substrate types are represented by: S (screen), R (crushed rocks), and C (smooth cobbles)

[3/] Some fish had not completely absorbed their yolk.

Table 5. Analysis of variance test for eyed coho and chum salmon eggs to test for significant differences in eye diameter due to stocking density and water velocity.

Table 5A. Differences due to water velocity:

Stocking Density	Calculated F-Statistic[2/]	L. S. D.[4/] $t_{.025,171}\sqrt{2S^2/20}$	Water Velocities Which Produced Significantly Larger Eye Diameters
eggs/cm^2		mm	cm/hr
1.5	1.73		
2.4	12.49*	0.061	54,220
3.3	11.98*	0.054	54,220

Table 5B. Differences due to stocking density:

Water Velocity	Calculated F-Statistic[3/]	L. S. D.[4/] $t_{.025,171}\sqrt{2S^2/20}$	Stocking Densities Which Produced Significantly Larger Eye Diameters
cm/hr		mm	eggs/cm^2
13	4.07*	0.069	1.5
54	5.89*	0.050	no evident trend
220	2.56*	0.054	no evident trend

* F greater than $F_{\alpha} = .05$

1/ The sample contained about 90% coho eggs and about 10% chum eggs.

2/ Treats all eye diameter measurements under each density, from all substrate types, and at all velocities.

3/ Treats all eye diameter measurements under each velocity, from all substrate types, and at all densities.

4/ L. S. D. = Least significant difference between any two pairs of eye diameter measurements.

Table 6. Analysis of variance test for coho alevins reared on a crushed rock substrate to test for significant differences in eye diameter due to stocking density and water velocity.

Table 6A. Differences due to water velocity:

Stocking Density	Calculated F-Statistic[1/]	L. S. D.[3/] $t_{.025,57}\sqrt{2S^2/20}$	Water Velocities Which Produced Significantly Larger Eye Diameters
eggs/cm^2		mm	cm/hr
1.5	44.34*	0.048	54,220
2.4	8.48*	0.070	54,220
3.3	32.87*	0.060	220

Table 6B. Differences due to stocking density:

Water Velocity	Calculated F-Statistic[2/]	L. S. D. $t_{.025,57}\sqrt{2S^2/20}$	Stocking Densities Which Produced Significantly Larger Eye Diameters
cm/hr		mm	eggs/cm^2
13	2.30		
54	4.53*	0.060	1.5, 2.4
220	3.01		

* F greater than $F_\alpha = .05$.

1/ Treats all eye diameter measurements under each density and at all velocities.

2/ Treats all eye diameter measurements under each velocity and at all densities.

3/ L. S. D. = Least Significant Difference between any two pairs of eye diameter measurements.

Table 7. Analysis of variance test for coho fry to test for significant differences in eye diameter due to substrate type, stocking density, and water velocity.

Table 7A. Differences due to substrate (crushed rock and smooth cobble):

Stocking Density	Water Velocity	Calculated F-Statistic	L. S. D. $t_{.025,38}\sqrt{2S^2/20}$	Substrate Which Produced Significantly Larger Eye Diameters
eggs/cm^2	cm/hr		mm	
1.5	13	1.94		
1.5	54	2.00		
1.5	220	0.29		
2.4	13	5.39*	0.067	smooth cobble
2.4	54	2.63		
2.4	220	0.68		
3.3	13	0.65		
3.3	54	1.86		
3.3	220	0.75		

Table 7B. Differences due to water velocity:

Stocking Density	Calculated F-Statistics[1/]	L. S. D.[3/] $t_{.025,114}\sqrt{2S^2/20}$	Water Velocities Which Produced Significantly Larger Eye Diameters
eggs/cm^2		mm	cm/hr
1.5	16.22*	0.052	220
2.4	16.56*	0.060	54, 220
3.3	7.72*	0.058	220

Table 7C. Differences due to stocking density:

Water Velocity	Calculated F-Statistics[2/]	L. S. D. $t_{.025,114}\sqrt{2S^2/20}$	Stocking Densities Which Produced Significantly Larger Eye Diameters
cm/hr		mm	eggs/cm^2
13	6.06*	0.063	no evident trend
54	4.47*	0.051	1.5, 2.4
220	5.97*	0.056	1.5

* F greater than $F_{\alpha} = .05$

1/ Treats all eye diameter measurements under each density, from all substrate types, and at all velocities.

2/ Treats all eye diameter measurements under each velocity, from all substrate types, and at all densities.

3/ L. S. D. = Least Significant Difference between any two pairs of eye diameter measurements.

COHO:

Stocking Density (eggs/cm^2)	Water Velocity (cm/hr) 13	54	220
1.5	▨	▨	▨
2.4		▨	▨
3.3		▨	▨

Eyed Eggs

Stocking Density (eggs/cm^2)	Water Velocity (cm/hr) 13	54	220
1.5		▨	▨
2.4			▨
3.3			▨

Alevins

Stocking Density (eggs/cm^2)	Water Velocity (cm/hr) 13	54	220
1.5			▨
2.4			
3.3			

Fry

CHINOOK: No Samples Taken

Stocking Density (eggs/cm^2)	Water Velocity (cm/hr) 13	54	220
1.5		▨	▨
2.4			
3.3			

Eyed Eggs

Stocking Density (eggs/cm^2)	Water Velocity (cm/hr) 13	54	220
1.5	▨	▨	▨
2.4		▨	▨
3.3			▨

Alevins

Stocking Density (eggs/cm^2)	Water Velocity (cm/hr) 13	54	220
1.5			
2.4			
3.3			

Fry

Figure 9. Production, from eyed-eggs to fry, of significantly larger (shaded areas) coho and chinook salmon reared under various stocking densities and water velocities.

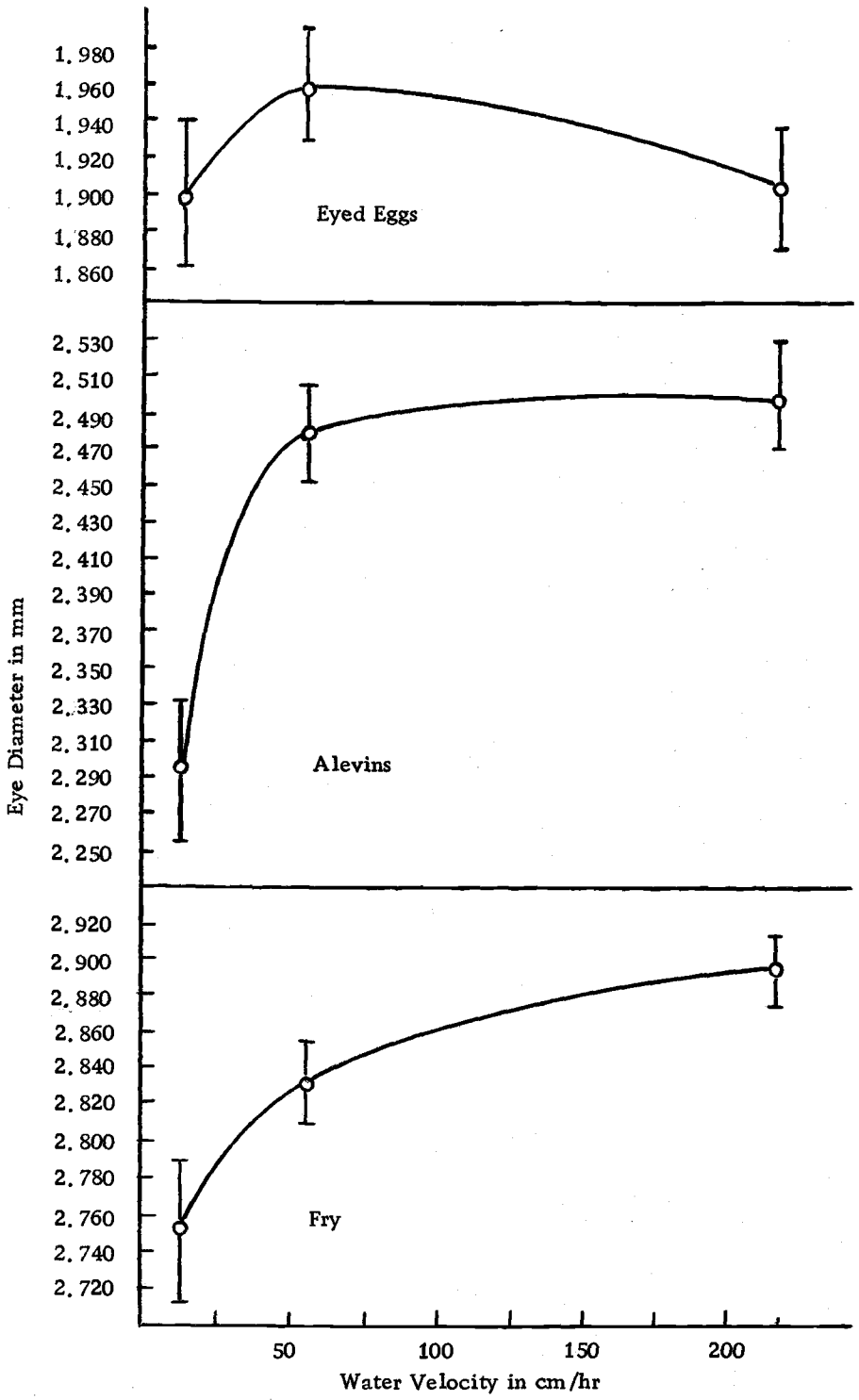

Figure 10. Influence of water velocity on the eye diameter of coho salmon (reared at low density and on a crushed-rock substrate) from eyed-eggs to fry. Vertical line through each point indicates the 90% confidence interval.

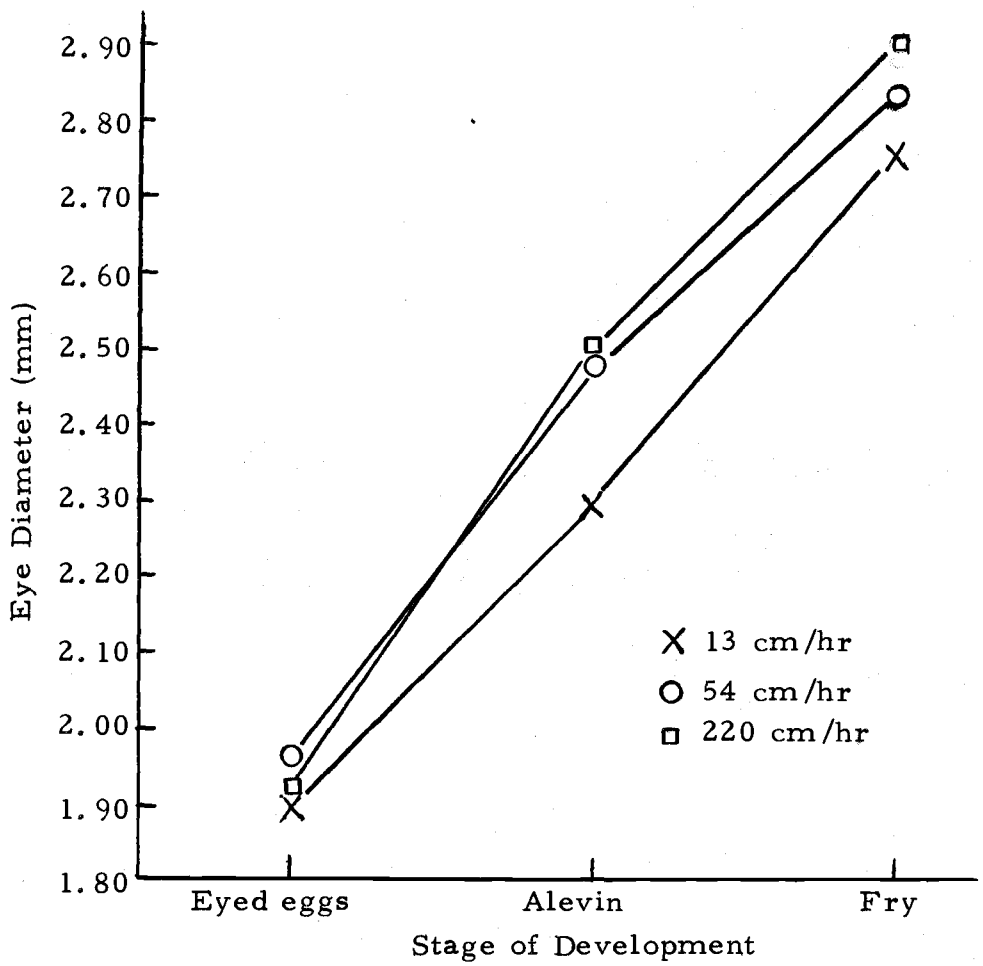

Figure 11. The increase of eye diameter of coho salmon through three developmental stages as influenced by three water velocities. The coho salmon were reared at low density and on a crushed rock substrate.

Substrate type produced a difference in downward movement of alevins, in time of fry emergence, and in reaction of fry to humans. After hatching, alevins readily penetrated into smooth cobbles. Those remaining on the substrate surface were quiescent and attempted to penetrate into the substrate when exposed to light. Swimming fry first appeared in cells with the screen substrate and last appeared in the cells with smooth cobbles. Low density and high water velocity also delayed the appearance of fry. Fry from smooth cobbles exhibited a greater fright reaction when disturbed than those from the screen and rock substrates.

The significance of the difference in alertness on subsequent survival is not known, but the difference in behavior may result from the environmental experience during early development. The alevins which remained above the screen and crushed rock substrates were exposed intermittently to light whenever the cover was lifted to inspect the cells; whereas, the alevins on the smooth cobble substrate moved downward where they were able to avoid light until emergence. Brannon (1965) observed a similar insensitivity in sockeye salmon exposed to light. Significantly, the more alert late swimmers were also the fry showing the most favorable growth.

Chinook Salmon in Test Cells

The chinook salmon eggs hatched about one week after their

introduction and mortalities remained low through the fry stage. They attained good growth under a wider range of velocities and stocking densities than coho salmon. Measurements of samples at the alevin and fry stages are presented in Table 8.

The F statistics (Tables 9 and 10) showed that crowding of chinook salmon had a greater influence than water velocity on growth to the alevin stage, but this difference was less evident to the fry stage. The best growth was initially afforded at the alevin stage by the low density cells with water velocities of 54 and 220 cm/hr. By the fry stage, fish exhibited good growth in all cells except for where velocity was 13 cm/hr in cells stocked at medium and high density and where velocity was 54 cm/hr in cells stocked at high density (Figure 9). This may have been due to an innate characteristic of the species or to the incubation of the eggs to the eyed stage in a Heath incubator before their introduction to the test cells.

The behavior of chinook salmon alevins and fry was similar to that observed in coho salmon.

Stocking of Eyed vs. Freshly-Fertilized Coho Eggs

When results of this experiment (Table 11) were compared with experimental cells in the larger experiment at similar stocking density, water velocity, and substrate type, no significant differences were observed in survival, hatching time, behavior, or growth,

Table 8. Eye diameter[1/] and total length[1/] (both in millimeters) of chinook salmon reared from alevin stage to fry emergence at various water velocities and stocking densities, and with different substrate types.[2/]

Stage of Development	Stocking Density	Water Velocity									
		13 cm/hr				54 cm/hr				220 cm/hr	
	eggs/cm^2	Eye Diameter		Total Length		Eye Diameter		Total Length		Eye Diameter	Total Length
ALEVINS	1.5	R	3.118±.027	R	37.40±.36	R	3.155±.022	R	37.40±.43	R 3.186±.021	R 37.85±.36
	2.4	R	3.078±.016	R	36.45±.36	R	3.088±.015	R	36.75±.33	R 3.098±.030	R 36.45±.46
	3.3	R	3.074±.018	R	36.15±.43	R	3.099±.023	R	36.50±.34	R 3.106±.026	R 36.50±.46
FRY[3/]	1.5	R	3.199±.026	R	39.55±.39	R	3.190±.023	R	38.55±.43	R 3.222±.030	R 38.75±.60
	1.5	C	3.215±.030	C	39.00±.33	C	3.182±.025	C	38.50±.51	C 3.257±.025	C 38.45±.43
	2.4	R	3.171±.028	R	38.00±.55	R	3.201±.019	R	39.05±.39	R 3.231±.016	R 39.20±.50
	2.4	C	3.176±.015	C	38.35±.32	C	3.213±.020	C	38.55±.50	C 3.246±.022	C 39.05±.43
	3.3	R	3.159±.029	R	38.05±.58	R	3.176±.016	R	38.95±.41	R 3.196±.021	R 39.30±.58
	3.3	C	3.155±.037	C	37.80±.57	C	3.154±.028	C	37.70±.45	C 3.203±.024	C 38.65±.41

[1/] Each is a mean of 20 measurements and is given with its 90% confidence interval.

[2/] Substrate types are represented by: R (crushed rocks) and C (smooth cobbles).

[3/] Some fish have not completely absorbed their yolk.

Table 9. Analysis of variance test for chinook alevins reared on a crushed rock substrate to test for significant differences in eye diameter due to stocking density and water velocity.

Table 9A. Differences due to water velocity:

Stocking Density	Calculated F-Statistic[1/]	L. S. D.[3/] $t_{.025,57}\sqrt{2S^2/20}$	Water Velocities Which Produced Significantly Larger Eye Diameters
eggs/cm^2		mm	cm/hr
1.5	6.18*	0.037	54, 220
2.4	0.64		
3.3	1.82		

Table 9B. Differences due to stocking density:

Water Velocity	Calculated F-Statistic[2/]	L. S. D.[3/] $t_{.025,57}\sqrt{2S^2/20}$	Stocking Densities Which Produced Significantly Larger Eye Diameter
cm/hr		mm	eggs/cm^2
13	4.30*	0.033	1.5
54	9.55*	0.032	1.5
220	10.31*	0.042	1.5

* F greater than $F_{\alpha} = .05$.

1/ Treats all eye diameter measurements under each density and at all velocities.

2/ Treats all eye diameter measurements under each velocity and at all densities.

3/ L. S. D. = Least Significant Difference between any two pairs of eye diameter measurements.

Table 10. Analysis of variance test for chinook fry to test for significant differences in eye diameters due to substrate type, stocking density, and water velocity.

Table 10A. Differences due to substrate (crushed rock and smooth cobble):

Stocking Density	Water Velocity	Calculated F-Statistic	L. S. D.[3/] $t_{.025,38}\sqrt{2S^2/20}$	Substrates Which Produced Significantly Larger Eye Diameters
eggs/cm^2	cm/hr		mm	
1.5	13	0.47		
1.5	54	0.33		
1.5	220	2.43		
2.4	13	0.00		
2.4	54	0.50		
2.4	220	0.98		
3.3	13	0.00		
3.3	54	1.38		
3.3	220	0.38		

Table 10B. Differences due to water velocity:

Stocking Density	Calculated F-Statistics[1/]	L. S. D.[3/] $t_{.025,114}\sqrt{2S^2/20}$	Water Velocities Which Produced Significantly Larger Eye Diameters
eggs/cm^2		mm	cm/hr
1.5	3.10*	0.042	no evident trend
2.4	6.40*	0.033	54, 220
3.3	1.90		

Table 10C. Differences due to stocking density:

Water Velocity	Calculated F-Statistic[2/]	L. S. D.[3/] $t_{.025,114}\sqrt{2S^2/20}$	Stocking Densities Which Produced Significantly Larger Eye Diameters
cm/hr		mm	eggs/cm^2
13	2.08		
54	2.64*	0.035	no evident trend
220	3.14*	0.037	no evident trend

* F greater F_{α} = .05.

1/ Treats all eye diameter measurements under each density, from all substrate types, and at all velocities.

2/ Treats all eye diameter measurements under each velocity, from all substrate types, and at all densities.

3/ L. S. D. = Least Significant Difference between any two pairs of eye diameter measurements.

Table 11. A comparison of eye diameter[1/] and total length[1/] (both in millimeters) from eyed eggs to emerged fry, of coho salmon reared entirely on a substrate versus those incubated to the eyed stage in a Heath incubator before introduction onto a substrate. All experimental cells were stocked at 1.5 eggs/cm^2 and water velocity was set at 54 cm/hr.

Stage of Development	Substrate Type	Rearing Method: Reared Entirely on Substrate		Rearing Method: Reared in a Heath Incubator Then on a Substrate	
		Eye Diameter	Total Length	Eye Diameter	Total Length
EYED EGGS	screen	1.899±.034	No Measurements	1.926±.044[2/]	No Measurements
	crushed rock	1.959±.031		1.926±.044[2/]	
	smooth cobble	1.876±.041		1.926±.044[2/]	
ALEVINS	screen	Data discarded	Data discarded	2.400±.038	27.80±.50
	crushed rock	2.478±.027	28.40±.29	2.523±.021	29.20±.34
	smooth cobble	No measurements	No measurements	No measurements	No measurements
FRY[3/]	screen	Data discarded	Data discarded	2.810±.024	34.68±.44
	crushed rock	2.832±.022	34.95±.32	2.827±.022	35.78±.33
	smooth cobble	2.804±.025	34.45±.51	2.805±.025	34.50±.41

[1/] Each is a mean of 20 measurements and is given with its 90% confidence interval.

[2/] Eggs had just been transferred from Heath Incubator to experimental cells, therefore a single sample was taken from all three substrates.

[3/] Some fish had not completely absorbed their yolk.

suggesting that substrate has no influence on growth and development. Substrate appeared to affect behavior of alevins and fry as previously described.

Comparisons Between Fish Held in the Heath Incubator with Fish From the Experimental Cells

The comparative data (Table 12) suggest that under the best conditions of water velocity (54 or 200 cm/hr) and stocking density (low), the experimental cells may produce more robust fry than the Heath incubator. Coho fry exhibited the best growth in the experimental cells and had a larger average eye diameter than fry from the Heath incubator. Some experimental cells produced chinook fry with larger eye diameters and slightly longer total lengths than from the Heath incubator.

Table 12. A comparison of eye diameter and total length (both in millimeters) of coho and chinook salmon reared in a Heath incubator with those reared in the experimental cells.

Species	Stage of Development	Rearing Method			
		Experimental Cells		Heath Incubator	
		Eye Diameter[1/]	Total Length[1/]	Eye Diameter[2/]	Total Length[2/]
COHO	Eyed Eggs	1.775±.050 to 1.992±.015	No measurements	1.966±.024	No measurements
	Alevins	2.237±.043 to 2.501±.029	26.30±.38 to 28.70±.36	2.541±.020	29.80±.32
	After Fry Emergence	2.585±.054 to 2.895±.021	32.90±.38 to 35.70±.75	2.860±.019	36.83±.29
CHINOOK	Alevins	3.074±.018 to 3.186±.021	36.15±.43 to 37.85±.36	3.130±.020	38.00±.43
	After Fry Emergence	3.154±.028 to 3.257±.025	37.70±.45 to 39.55±.39	3.210±.026	38.35±.53

1/ The lowest and the highest measurements (each a mean of 20 measurements and given with its 90% confidence interval) are given from all the experimental cells.

2/ Each is a mean of 20 measurements and is given with its 90% confidence interval.

OPERATION OF A PROTOTYPE STREAMSIDE INCUBATOR

The prototype streamside incubator (Figures 12, 13, and 14) was constructed at Netarts Bay (Figure 15) in summer 1968. Water was pumped to the hatchery from Whiskey Creek. This account considers the period of operation ending in April 1969.

Design and Operation

The hatchery is located 300 feet upstream from the mouth of Whiskey Creek. A one-horsepower pump delivered water at about 30 gallons per minute to two head tanks. Water flowed by gravity from the head tanks to 12 hatchery tanks arranged in four units each consisting of three interconnecting tanks (see Figure 16). Water downdrafted past the eggs through the substrate and was collected by an underdrain system and discharged through outflow standpipes (see Figure 17 for schematic diagram of hatchery tank). Rate of the outflow and the depth of the water layer was controlled by adjusting the height of the standpipes. The hatchery tanks were covered to exclude light.

A commercial sand-filter was installed between the head tanks and the hatchery tanks. Operational problems precluded its use, however.

Figure 12. Prototype hatchery as seen from road.

Figure 13. Front view of prototype hatchery.

Figure 14. Rear view of prototype hatchery.

Figure 15. Netarts Bay, Oregon

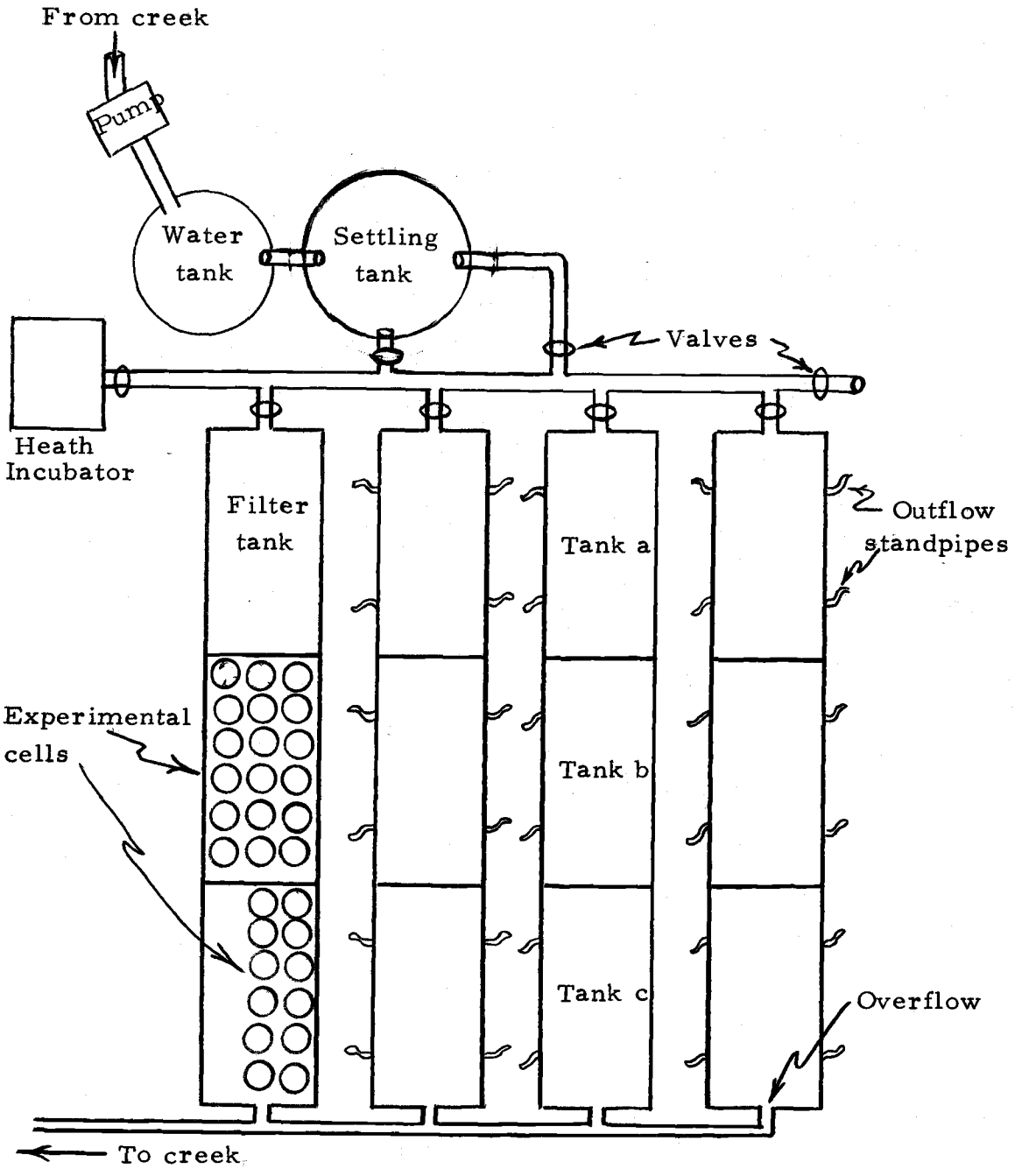

Figure 16. Schematic top view of prototype hatchery showing water delivery system.

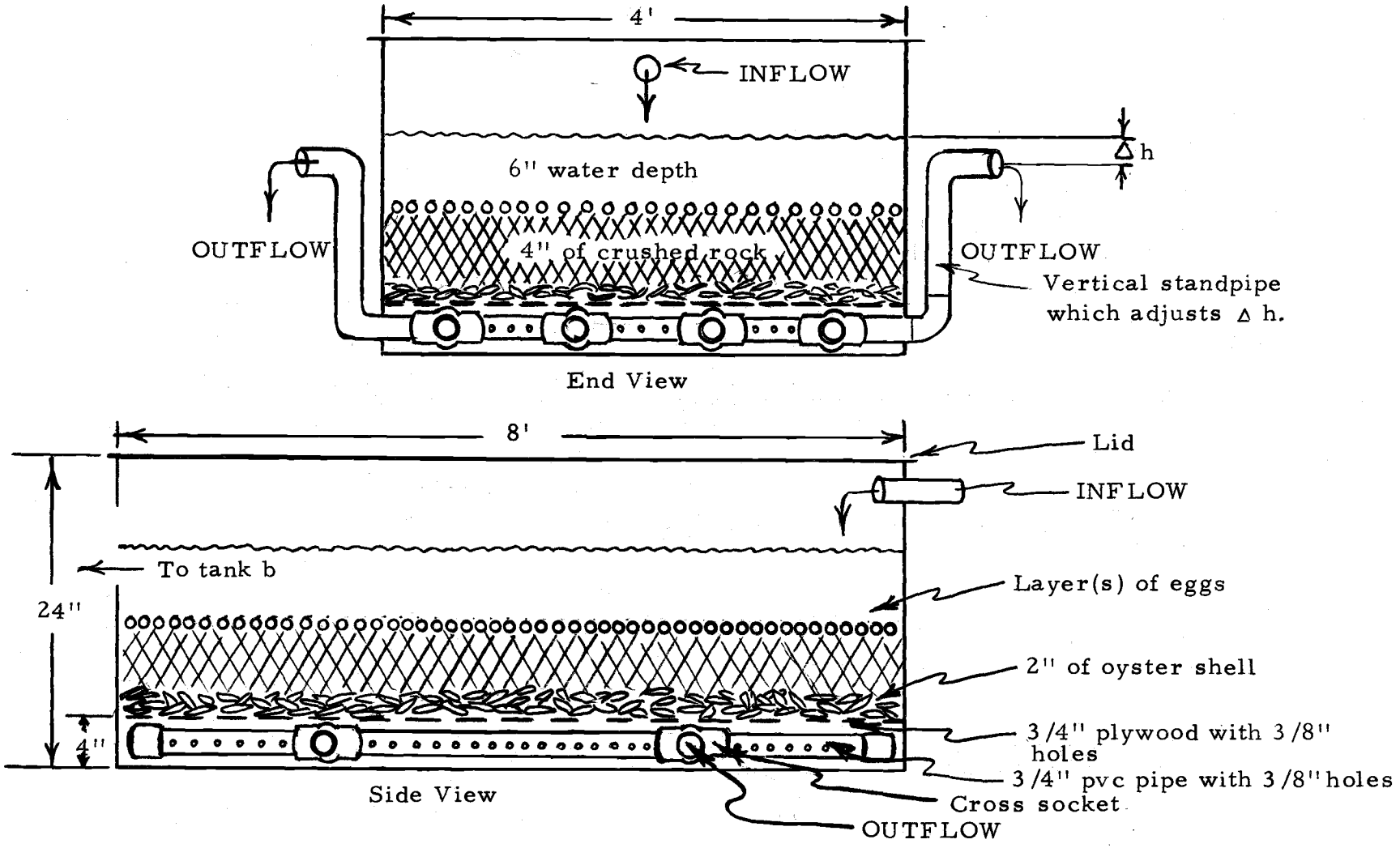

Figure 17. End and side view of hatchery tank a showing the direction of water flow.

Based on the preliminary experiments conducted during 1967-68, the prototype was thought to have a capacity for at least 1.2 million eggs reposing on 382 ft^2 area of crushed rock substrate (three eggs per cm^2 or about 3000 per ft^2). A layer of oyster shell was provided between the crushed rock and the underdrain system to disperse the flow of downdrafting water across the surface of the bed. Water velocity past the eggs and alevins was initially planned for 25-50 cm/hr; however, decreased pump efficiency and demands for water for designed experiments allowed only enough water for a velocity of 15 cm/hr during winter and spring 1969.

The eggs were treated with malachite green twice weekly until hatching to reduce the growth of fungus on dead eggs.

At the time of fry emergence and migration, water flow through the underdrain was reduced to allow water to pass through the common overflow at the end of each unit of three tanks; the intention was to provide fry free access to Netarts Bay without handling them.

Stocking

Four species of Pacific salmon - pink, coho, chinook, and chum, and two hybrid salmon - were used in the first year to test the hatchery and to conduct the experiments described elsewhere in this thesis. The eggs came from several sources in Alaska, Oregon, and Washington. Some were taken from wild stocks and others from

hatchery stock. The methods used in preparing the eggs for shipment also varied considerably; some were shipped unfertilized, some were shipped fertilized, and some were shipped eyed. Mortality from handling varied greatly amongst the several groups introduced into the hatchery.

Pink salmon eggs were acquired in Alaska on September 15, 1968, to initiate tests on the system. These eggs were held at the Oregon Fish Commission Big Creek Hatchery in Astoria, until the hydraulic characteristics of the prototype incubator could be evaluated. On October 22, an estimated 300,000 eyed pink salmon eggs were transported from Astoria to Netarts Bay and placed in three hatchery tanks.

Operational hazards were encountered soon after the first pink salmon eggs arrived from the Big Creek Hatchery; the water pump was inundated by a 10-foot tide in mid afternoon of October 22. We installed a gasoline pump temporarily near the intake box until the electric pump could be relocated on higher ground near the hatchery. In early November, a tree struck the intake box, and a strong freshet on November 9 dislodged the box completely, causing a water stoppage which could have lasted 14 hours before repairs could be made. This stoppage caused all but 12,000 of the original pink salmon eggs introduced to the hatchery to suffocate.

Because of uncertainties about the reliability of the water

system, native chum salmon were not taken from Whiskey Creek in late November as originally planned. Instead, surplus coho eggs were obtained from the Oregon Fish Commission Trask River Hatchery near Tillamook and surplus chinook eggs from the University of Washington. About 110,000 coho eggs were introduced in late November and 100,000 chinook eggs in early December. Five hatchery tanks were used for these fish.

Experimental cells (described earlier) were also set up in two hatchery tanks to investigate the effects of stocking density, water velocity, and substrate type; 44,000 coho eggs, 6,000 chinook eggs, and 6,000 chum eggs were used in these experiments.

Hybridization was attempted with one group of 16,000 coho eggs by fertilizaing them with chum salmon sperm. Initial fertility (after 20 days) was estimated at 70 percent; however, the embryos were deformed, and none survived to hatch. A viable hybrid salmon was produced by crossing eggs from a chum salmon with sperm from a chinook salmon; the resulting fish was called a "chumook." About 1,000 chumooks were brought from the Fish Commission Big Creek Hatchery to Netarts Bay where they hatched and developed into normal appearing fry. These fish were later transferred to the Port Orford Fisheries Laboratory for sea water acclimation experiments.

The six hatchery tanks of the prototype streamside incubator used in the first year produced approximately 12,000 pink, 8,000

coho, and 75,000 chinook salmon. In addition, about 20,000 coho and several thousand chum and chinook salmon were produced in the test cells. About 7,000 pink salmon were released into Netarts Bay between January 6 and February 6. The remainder of the fish were used in experiments at Port Orford, Corvallis, and Newport.

The initial egg stock included 300,000 pink, 110,000 coho, and 100,000 chinook salmon; hence mortality exceeded survival. An estimated 288,000 pink salmon eggs were lost due to the water stoppage in early November; 33,000 newly fertilized coho salmon eggs were known to have died as a result of rough handling; and 16,000 coho eggs were included in the unsuccessful attempt at hybridization with chum salmon. Other causes of mortality will be discussed in the next section.

Operational Problems

Most operational problems were attributable to an undependable pumping system, leakage of tanks, and poor water circulation within the tanks.

The Pumping System

The intake box in the creek was vulnerable to dislodgment, which occurred once on November 9 (the replacement remained to the end of the season). Gravel covering the intake box provided little filtration;

pebbles, twigs, and other small debris had to be cleaned frequently from the pump impeller. In the absence of a suitable filtration system, silt entered the hatchery during each heavy rain.

Wild animals occasionally inflicted damage on the water line. Periodic repairs were necessary to avoid air in the suction line. By January, 1969, the pump started to suck air, causing a gradual decline in delivery capacity. As a result, water velocity in the hatchery tanks had to be decreased from about 25 cm/hr to 15 cm/hr.

Power outages were common, but the hatchery tanks were designed to retain standing water; an outage exceeding six hours, however, can cause serious problems from oxygen depletion. Generally, outages lasted less than two hours, a period tolerable under normal weather conditions (air temperature ~50°F and water temperature ~45°F). A backup gasoline pump was not available at the hatchery until March 1969.

Weather and Tidal Conditions

The winter of 1968-69 was the worst recorded in 20 years. Air temperatures declined to 18°F and water temperatures to near freezing. Snow covered the hatchery in December and throughout January; but freezing weather caused no operational difficulties so long as the pump was operating efficiently. Frozen water lines almost resulted on December 20 when a power outage lasted two hours at a time when the

air temperature dropped to 18°F. The cold water in December and January had no detrimental effect on the salmon.

The highest tides came to within two feet of the base of the hatchery. The pump was inundated during the first high tide of autumn. Relocation to higher ground corrected this problem.

Leakages

The seams of the hatchery tanks were not water-tight and leakages had to be sealed with wet patch and oiled fibers (known commonly as horse hair). In one instance, a leakage was serious enough to lower the water level in one series of three hatchery tanks.

Poor Circulation in Hatchery Tanks

Egg mortality in the hatchery tanks was partly attributable to inadequate water velocity stemming from four factors: high egg density, low water volume from reduced pump efficiency, fungus on dead eggs, and silt.

Eggs were intended to be distributed within the hatchery tanks as a single layer. Spreading of a uniform layer of eggs proved to be difficult, however, and pockets of low and high density resulted.

Because of decreased efficiency of the pump in early January 1969, it became necessary to reduce water velocity in the hatchery tanks from 25 to 15 cm/hr. This low velocity may have been

insufficient for the removal of waste metabolites and the resupply of dissolved oxygen to some of the eggs.

Eggs became covered with a film of silt and mortality was difficult to assess immediately. It was also extremely difficult to remove the dead eggs from the rock substrate. Dead eggs were infected by fungus, which further restricted the flow of water. Where there was early mortality from handling, dead eggs could not be removed without risking further mortality of adjacent, uneyed eggs. In some instances high mortality took place all at once just before hatching, and the removal of dead eggs became a difficult and time consuming task. In many cases, silt combined with the dead eggs and fungus to form a solid mat of material, choking off the water circulation.

The consequence of the poor invironment was dramatically demonstrated in one hatchery tank where 16,500 eggs (coho) with an estimated initial fertility of 91 percent produced less than 50 percent survival at hatching. By inspection, mortalities were mostly eyed eggs, where they had become concentrated in more than one layer.

Biological Observations

Conditions in the hatchery tanks were least favorable for the eggs which were introduced soon after fertilization. Survival of coho eggs incubated from fertilization was less than 50 percent at hatching.

Chinook eggs, which had been introduced as eyed eggs, had at least 75 percent survival at hatching. The survival of alevins was high for all species, probably because of their self aeration ability (Bams, 1969).

A wide difference in rate of development was observed within each hatchery tank; hatching was spread out over long periods and alevins were still present in the rock substrate after many free-swimming fry had emerged.

The observed behavior of pink, coho, and chinook alevins and fry was basically the same. After hatching, alevins repose quietly on the substrate, becoming active and seeking cover only when exposed to light. Before yolk absorption was completed and neutral buoyancy was attained, light elicited either a digging response or a swim-up response of short duration. After the fish had attained neutral buoyancy, light elicited schooling and circular swimming patterns within the tanks, generally opposite to the current.

Several thousand pink salmon fry were able to penetrate through the crushed rock and enter the underdrain system. Many of these appeared in the outflow but others became trapped and died. Coho and chinook salmon alevins did not or were not able to penetrate in great numbers downwards into the crushed rock. The rock was 1/4 to 1/2 inch diameter.

Deformed fry were not observed in sizeable numbers. Whirling

was the most prevalent abnormal behavior but it was observed in less than one percent of the population.

Suggested Design Improvements

A number of improvements in the system are necessary to increase its reliability:

1) Remove intake system from the creek. This would reduce the possibility of the intake being washed out. It would also reduce damage to the suction line from animals. A settling pond is scheduled for construction next to Whiskey Creek. The plan is to divert water through a culvert into the settling pond before it is pumped into the hatchery.

2) Provide a filtration system to eliminate silt. Silt caused reduced circulation within the hatchery tanks. Logging on the Whiskey Creek watershed will continue to muddy the stream during freshets, and an upwelling filtration system is suggested. The first tank in each series of three tanks could be converted for this purpose.

3) Increase water velocity in hatchery tanks. Results of the experiments on water velocity show that velocities of 50 to 100 cm/hr provide better growth and survival than lower velocities. Hatchery tanks should be redesigned to maximize water velocity with a given inflow volume by circulating all of the water through each tank in series before discharging the used water.

4) Provide a backflush system. No suitable system for backflushing the incubation tanks existed in the prototype. A backflush system is needed when circulation becomes impaired.

5) Elevate eggs off substrate. Eggs should be incubated on screens rather than on a rock substrate to facilitate the assessment and removal of mortalities and to avoid clustering. The screen could retain the eggs until they hatch and allow the alevins to fall freely onto the rock substrate to complete their development.

6) Provide a coarse rock substrate (or its equivalent) in which the alevin can hide. A coarse rock substrate not only produced a more alert fry in the designed experiments, but also provided the developing alevins better opportunities to hide and avoid light.

7) Have an auxiliary source of power. A backup gasoline pump should be hooked up and ready to go in case of mechanical failure of the electric pump or in case of a prolonged power outage.

8) Eliminate leaks from the hatchery tanks. This can be done with better construction and by giving the seams a better sealing.

9) Have a hatcheryman in residence. Many of the problems encountered could have been avoided if they had been detected without delay.

TRANSPORTATION OF FRESHLY SPAWNED EGGS

Previous research on delayed fertilization of salmonid eggs has dealt mostly with the storage of small quantities of gametes which were subsequently crossed with fresh gametes. High fertility has been obtained for sockeye salmon where sperm had been stored in contact with air for periods up to 24 hours and ova up to 96 hours (Foerster, 1965). Studies with sockeye and pink salmon (Withler and Humphreys, 1967; Winthler and Morley, 1968) showed that cooler temperatures prolonged the period over which stored gametes would remain viable; at 3°C, the lowest temperature used, both ova and milt of pink salmon retained their initial high fertility for 46 hours.

Unless fresh milt or eggs are available at the destination, the practical application of delayed fertilization in transplanting eggs will involve their fertilization with stored sperm (Barrett, 1951); with few pink salmon spawning in Oregon, our transplant is an example of such a situation. Unfortunately, delayed fertilization with stored gametes has received little attention.

To prepare for the transplant, pilot studies were carried out in summer 1968 at the Bureau of Commercial Fisheries' Little Port Walter Research Station, Baranof Island, southeastern Alaska. The transplant was conducted on September 15 and two subsequent field tests were carried out in Oregon in October and November to further evaluate the techniques used in the transplant. The individual

experiments are described chronologically.

Experiments at Little Port Walter, Alaska

The experiments at Little Port Walter provided information on the period within which acceptable fertility can be expected from stored gametes. They also provided experience on methods to be used in collecting and storing gametes for shipment.

Gametes were taken from pink and chum salmon spawners in Lovers Cove Creek near Little Port Walter. Adults were sorted, killed, and excess mucous and moisture were removed before the sex products were extracted. Sperm was collected with a sterilized dual tube suction apparatus (Horton, Graybill and Wu, 1967) (Figure 18) and inserted into 2-liter plastic jars. Ova were stripped by hand and placed into plastic whirl-paks (about 400 ml capacity). The gametes were transported five miles by boat to Little Port Walter and placed in a refrigerator. Storage temperatures varied between 4 and 9°C and averaged 6°C. Some test groups were shaken periodically to stimulate motion in transit.

Two types of storage experiments were run. With the first type, ova and milt were stored for 72 hours and then crossed with the fresh and/or stored gametes. Table 13 summarizes the results with stored eggs and Table 14 summarizes the results with stored milt. With the second type, stored ova and milt were mixed with fresh and/or stored

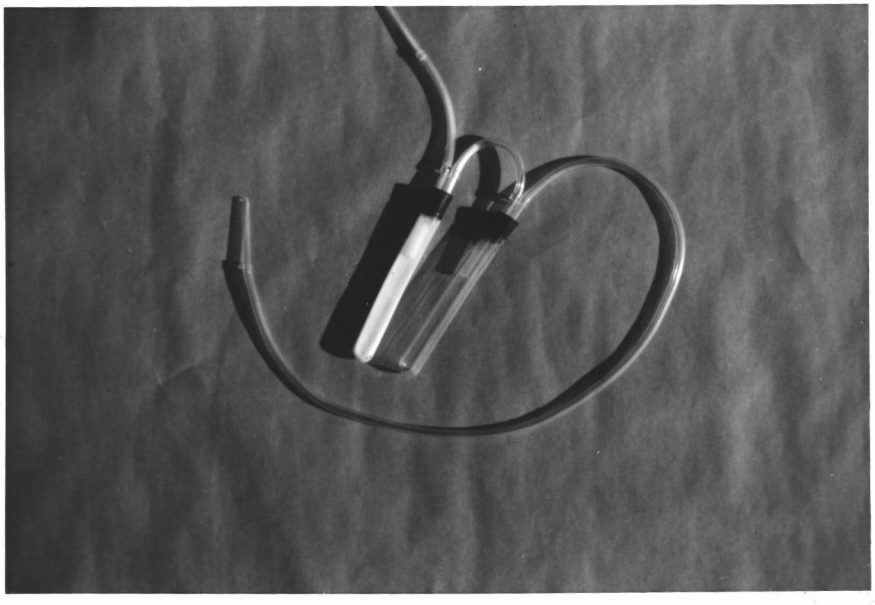

Figure 18. Dual tube suction apparatus used to collect sperm.

Table 13. Delayed fertilization experiments with pink and chum salmon eggs stored 72 hours.

Species	Quantity of Eggs	Ratio of Volume of Eggs to Air	Mechanical Agitation	Condition of Milt	Method of Adding Milt	Number of Eggs Examined Microscopically	Eggs Undergoing Cleavage
							percent
CHUM	1200[3/]	2:1	None	Fresh	dry[1/]	34	100
				Stored for 72 hours	diluted[2/]	65	46
	800[3/]	1:1	None	Stored for 72 hours	dry	74	31
				Fresh	dry	45	96
PINK	2250	2:1	None	Fresh	dry	72	82
	750	1:2	Shaken Every 2 Hours Approximately	Fresh	dry	57	93

1/Milt and eggs were mixed before water is added.

2/Milt was diluted with water before mixing with eggs.

3/One experiment; eggs were from a common lot.

Table 14. Delayed fertilization experiments with stored pink and chum salmon milt stored for 72 hours and mixed with fresh eggs.

Species	Quantity of Milt	Ratio of Volume of Milt to Air	Mechanical Agitation	Method of Adding Milt	Number of Eggs Examined Microscopically	Eggs Undergoing Cleavage
	milliliters					percent
PINK	60	1:30	shaken periodically	dry[1/]	53	43
PINK	30	1:70	shaken periodically	dry	51	39
				diluted[2/]	53	72
PINK	60	1:30	shaken periodically	dry	45	40
				diluted	56	80
CHUM	50[3/]	1:10	none	dry	54	20

[1/] Milt and eggs were mixed before water is added.

[2/] Milt was diluted with water before mixing with eggs.

[3/] Contaminated with urine, feces, blood and slime.

gametes at 6-hour intervals up to a total of 72 hours. The resulting curves relating fertility to time in storage are shown in Figure 19. Whenever possible, control groups were fertilized at the time gametes were taken.

After a test group of stored gametes had been fertilized, embryonic development was allowed to proceed up to two days. The eggs were preserved in freshly mixed Carnoy's fixative (three parts 95 percent alcohol and one part glacial acetic acid) and blastodiscs were removed and examined under low magnification for evidence of cell division. Factors studied included:

1) Air in storage containers. The need for air space in gamete storage is still not completely understood. Withler and Morley (1968) suggested that practical application of delayed fertilization probably should permit some contact of gametes with air. Truscott et al. (1968) recommended that a large air interface be provided where sperm is stored.

In our experiments, eggs were stored at egg-to-air volume ratios of 2:1, 1:1, and 1:2, with egg-air interface of 50-100 cm^2. Milt was stored at milt-to-air volume ratio of about 1:10 (not varied in the study) with milt-to-air interface of about 120 cm^2.

The experiments revealed that air space exerted only a minor influence on fertility of ova. In two experiments where pink salmon ova stored for 72 hours were crossed with fresh sperm, the results

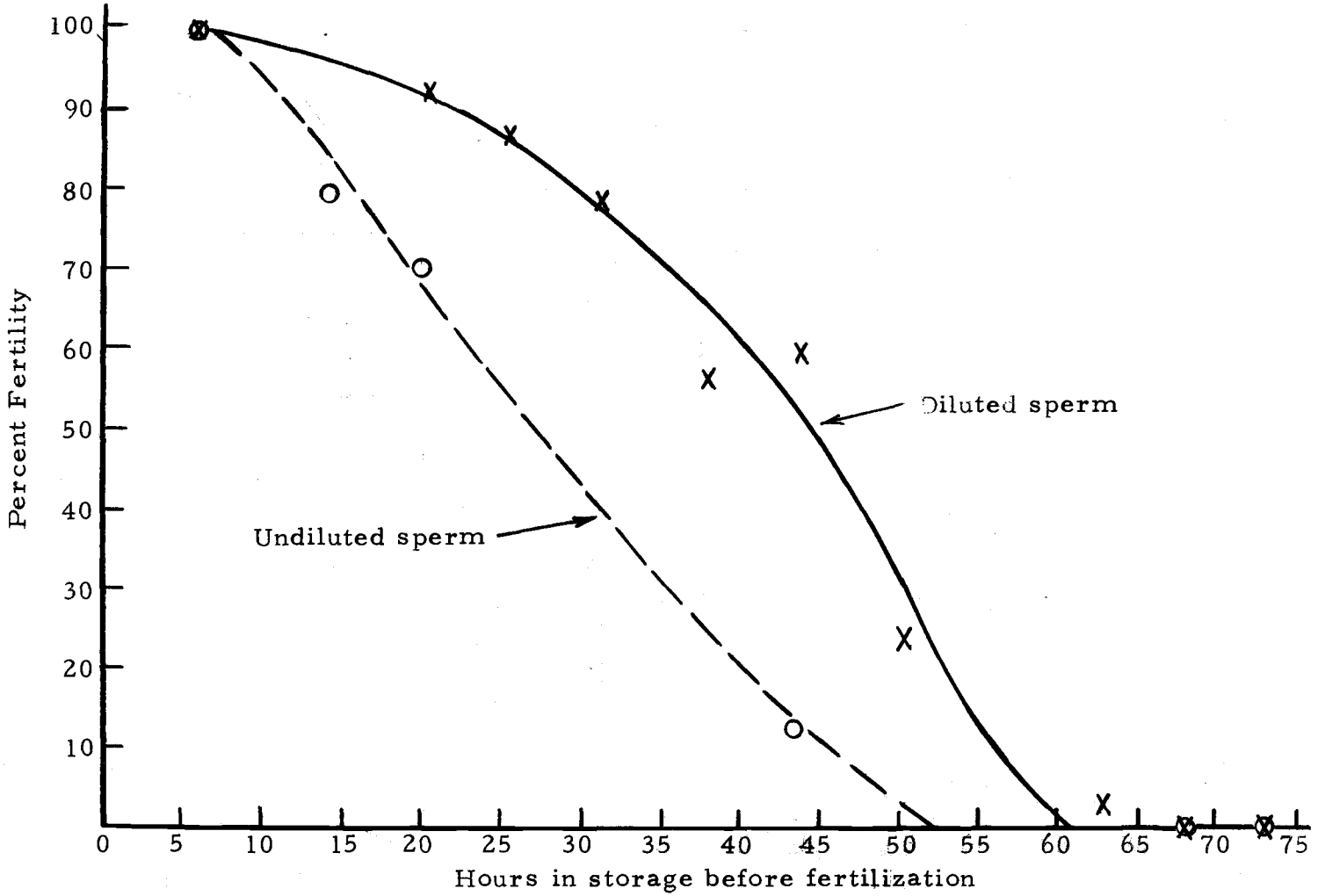

Figure 19. Percent fertilization of stored pink salmon ova fertilized with stored sperm. Storage temperature was 6°C.

suggest some possible effects due to air space:

Ova-to-air ratio	Percent fertilization
2:1	82
1:2	93

However, chum ova under the same conditions gave 96 and 100 percent fertility, suggesting that within a 72 hour period, the metabolism of unfertilized eggs is low enough such that a large air space is not needed.

2) Dilution of Sperm. In the course of one experiment, water was added to a packet of stored chum salmon sperm before it was mixed with stored ova. This diluted sperm gave 45 percent fertilization, which in this instance compared to only 31 percent for undiluted sperm (see Table 14). Followup experiments confirmed that dilution of stored sperm before mixing with ova resulted in higher fertility. Pink salmon gametes stored at 6°C gave 90 percent or higher fertility for periods up to 20 hours when the stored sperm was diluted with water before being mixed with stored ova. Nearly 100 percent fertility was possible for short periods up to six hours with or without addition of water.

3) Method of Gamete Extraction. Contamination by water, broken eggs, waste products, and slime were avoided in all spawning operations. Hand-stripping was slow and inefficient (about 1/3 of the eggs remain in the body cavity) but contamination was effectively avoided. Removal of milt by the suction apparatus is also slow, but

with proper execution, uncontaminated milt can be removed effectively. The suction tube should be inserted no more than one inch into the genital pore and at an angle of about 30 degrees from the long axis; shallow insertion allows for the draining of the junction of both seminal ducts and insertion at 30 degrees avoids the ureter opening.

4) Period of Fertility. When fresh gametes were crossed with gametes stored for 72 hours, pink salmon ova gave 82 to 100 percent fertility; whereas, pink salmon milt gave 72 and 80 percent fertility, even with the dilution method. Therefore, the quality of the milt appears to be the limiting factor in delayed fertilization. In every experiment, activation of stored sperm by water before mixing with the ova resulted in increased fertility.

5) Effects of Mild Agitation. Mild agitation seemed to have no effect on fertility. There nveretheless was concern that severe agitation, such as that experienced on a plane trip, might have deleterious effects.

Pink Salmon Transplant from Alaska to Oregon

Results of tests with stored gametes at Little Port Walter were sufficiently encouraging to attempt the movement of stored gametes from Alaska to Oregon. Pink salmon were spawned at Lovers Cove Creek in the morning of September 15, 1968, and gametes were packed

with ice in insulated (styrofoam) coolers. They were transported to Juneau aboard a Bureau of Commercial Fisheries amphibious aircraft; from Juneau to Seattle, Washington, via commercial jet; and from Seattle to Astoria, Oregon, by motor vehicle. The maximum time in storage was 18 hours; the minimum was 14 hours. (The duration of the spawning operation was four hours.)

An estimated 150,000 ova from about 100 female pink salmon were stripped into 1/2 gallon plastic jars. Each jar was filled to capacity with approximately 8,800 ova and accompanying fluid and sealed to exclude air.

Sperm was extracted from males and placed in 35 individual plastic packets, each containing 10 to 50 cc of sperm and 200 to 300 cc of air. Upon arrival at Astoria, sperm from two visually uncontaminated packets was combined and diluted with about 400 cc of water to fertilize eggs from one jar. The newly fertilized eggs were incubated at the Fish Commission of Oregon Big Creek Hatchery.

A second group of 250,000 pink salmon eggs was obtained and fertilized simultaneously with the collection of the unfertilized eggs. These were handled according to more typical hatchery practice and shipped along with the stored gametes. The fertilized eggs were washed and partially water hardened before being placed in commerical-type shipping trays lined with moist cheesecloth. Trays were placed in cardboard cases with ice.

The transplant provided an opportunity to test the need for an air space in the storage of ova. Four whirl-paks were filled with 300 cc of eggs (2,250 eggs and a 2:1 egg-to-air volume ratio) and four more were filled with 100 cc of eggs (750 eggs and a 1:2 egg-to-air volume ratio). These whirl-paks were placed in styrofoam coolers.

At the Big Creek Hatchery three out of four whirl-paks from each of the two air space ratios were fertilized in the same manner as the other delayed fertilization eggs. Nineteen hours later, after 37 hours of storage, the last whirl-pak was fertilized at the Marine Science Center in Newport, Oregon; a subsample of about 80 eggs was fertilized and incubated at 0°C.

After 16 days of incubation, subsamples of eggs were taken from all experimental groups and preserved in Carnoy's faxative. Fertilization was determined by the percentage of eggs which exhibited an embryonic streak.

Success of fertilization of the stored gametes was 92 percent, with little additional mortality through the eyed stage. The results of the transplant and the air space experiments are presented on Table 15. The group fertilized before shipment suffered 21 percent mortality during transit and an additional 20 percent during incubation to the eyed stage. Rough handling was evidenced by some trays which were damaged, and by the shifted position of eggs and cheese-cloth within the trays. The deleterious effect of mechanical shock on

Table 15. Results of pink salmon egg transplant,[1/] employing various handling methods, on 15 September 1968.

Type of Container	Number of Eggs Per Container	Ratio of Volume of Eggs to Air	Hours in Storage Before Fertilization	Number of Eggs Examined for Embryonic Streak	Percent of Eggs with Embryonic Streak
1/2 gallon jar[2/]	13,000	1: ~0	16	508	92
whirl-pak[4/]	2,250	2:1	16	109	93
whirl-pak[4/]	750	1:2	16	126	98
shipping tray[3/]			0	213	79
whirl-pak[4/]	2,250	2:1	37	77	95
whirl-pak[4/]	750	1:2	37	81	96

1/ From Lover's Cove Creek, Southeastern Alaska, to Big Creek Hatchery, Oregon.

2/ About 150,000 eggs were packed in this fashion and fertilized with stored milt at the end of 16 hours.

3/ About 250,000 eggs were fertilized at Lover's Cove, partially water-hardened, and placed in styrofoam shipping trays lined with moist cheese cloth.

4/ Subsidiary air space experiment.

fertilized eggs of pink salmon has been shown by Smirnov (1954). The difference in mortality between the two methods may be due to the difference in sensitivity of unfertilized and fertilized eggs to shock.

There was no effect on the ratio of ova-to-air on fertility. The observed values were:

Ova-to-air ratio	Percent fertilization
1:0	92
2:1	93
1:2	98

The differences among the sample means were not significant at the five percent level ($X^2 = 5.043$ with 2 d.f.).

By separating the milt from each male, contamination of the lot is avoided and quality milt can be isolated and used for fertilization. The dilution of sperm not only gave the expected high fertility (92 percent) based on the pilot studies (see Figure 19), but it made possible the fertilization of a large volume of eggs with a small quantity of milt.

Field Tests in Oregon

The pink salmon transplant demonstrated the possibility of transporting unfertilized gametes. Our results suggest that air space can be eliminated in storage of unfertilized ova, but other workers (Withler and Morley, 1968) have suggested that it be provided. Our methods of gamete extraction, though satisfactory, were slow; other

methods may suffice, i. e. stripping eggs by incision and milt by hand. Barrett (1951) stored a dry mixture of cutthroat trout eggs and milt for 24 hours and obtained 99 percent fertility; this method could be better than keeping gametes separate for delayed fertilization.

To investigate the above points, field tests were undertaken in October and November of 1968 to: 1) substantiate the delayed fertilization techniques used in the pink salmon transplant; 2) test other methods of gamete extraction; 3) test the cold storage of a dry mixture of eggs and milt as an alternative method of delayed fertilization.

Surplus coho eggs were obtained from the Oregon Fish Commission Big Creek Hatchery on October 17 and from the Trask River Hatchery on November 22. Eggs were stripped by incision or by hand and measured volumes were placed into plastic containers. Milt from each male was stripped by hand and divided between two whirl-paks. A small quantity of milt was then added dry to two containers of eggs. The gametes were stored in iced styrofoam coolers and transported by car to Netarts Bay where, at the end of 18 and 30 hours, delayed fertilization was carried out using the dilution method, and water was introduced to the dry mixture. During storage, the gametes were subjected to at least five hours of agitation by auto trips.

The fertilized eggs were incubated in a Heath incubator, and after 15 days, subsamples were preserved in Stockard's solution (four parts acetic acid, five parts formalin, six parts glycerin and

85 parts water). Fertilization was determined from the percentage of eggs with embryonic streaks.

At Big Creek, eggs were stripped by the incision method. Six plastic freezer containers (about 1,300 eggs per pint-sized container) were filled with eggs, leaving no air space. Four pints were used for two delayed fertilization experiments, and to each of the two remaining pints, several drops of milt were added and the mixture stirred with a dropper. Although the eggs were stripped with care, some slime, water, waste products, and blood were observed in the samples.

At the Trask River Hatchery, eggs were stripped by hand and placed into four whirl-paks, two with about 1,600 eggs (1:0 egg-to-air volume ratio) and two with about 400 eggs (1:2 eggs-to-air volume ratio). About 20 cc of milt was added to about one quart of eggs; the mixture was stirred and about 400 eggs (1:2 eggs-to-air) were placed into each of two whirl-paks.

The results of the two field tests are presented in Tables 16 and 17, and summarized below.

Contamination

Contamination resulting from the incision method probably caused the consistently low fertility in the Big Creek field test; uncontaminated eggs of the Trask River field test showed a 48 and 31 percent improvement in the delayed fertilization experiments (with a

Table 16. Results of the transplant of Big Creek Hatchery coho salmon eggs[1/] employing various handling methods, on 17 October 1968.

Method of Treatment	Number of Eggs Per Container	Ratio of Volume of Eggs to Air	Hours in Storage	Number of Eggs Examined for Embryonic Streak	Percentage Eggs with Embryonic Streak
Delayed fertilization	1900	1:~0	18	245	63
Delayed fertilization	1900	1:~0	30	249	68
Dry mixture of milt and eggs	1900	1:~0	18	320	42
Dry mixture of milt and eggs	1900	1:~0	30	217	49
Fertilized control			0	138	96
Unfertilized control			30	132	0

[1/] Eggs transported from Big Creek Hatchery to Netarts Bay.

Table 17. Results of the transplant of Trask River Hatchery coho salmon eggs[1/] employing various handling methods, on 22 November 1968.

Method of Treatment	Number of Eggs Per Container	Ratio of Volume of Eggs to Air	Hours in Storage	Number of Eggs Examined for Embryonic Streak	Percentage Eggs with Embryonic Streak
Delayed fertilization	1500	1:~0	19	201	83
Delayed fertilization	1500	1:~0	30	178	89
Delayed fertilization	400	1:2	19	102	75[2/]
Delayed fertilization	400	1:2	30	127	87
Dry mixture of milt and eggs	400	1:2	19	116	84
Dry mixture of milt and eggs	400	1:2	30	139	86

[1/] From Trask River Hatchery to Netarts Bay. Control was accidentally killed.

[2/] Broken eggs were present and coagulation of yolk material occurred as water and added.

1:0 eggs-to-air volume ratio) and a 100 and 76 percent improvement in the dry mixture experiments. The faster and more efficient incision method, though subject to addition of contaminants, may still be acceptable if more care were to be taken during egg taking.

Hand-stripped milt retained its fluid consistency after 30 hours of storage; this method seems satisfactory for delayed fertilization.

Delayed Fertilization with Coho Eggs

Under the same storage conditions (1:0 eggs-to-air volume ratio; about 16 hours of storage), the delayed fertilization experiments at Trask River produced slightly lower fertility (83 percent) than the pink salmon transplant from Alaska (92 percent). This difference, however, may be due to variations between experiments, a species difference, gamete quality, or the method of milt extraction (hand-stripping at Trask and suction at Alaska), none of which have been adequately compared.

Air Space in Storage

Air space was again uncritical in the cold storage of unfertilized ova; however, it did play a role in the dry mixture experiments.

Dry Mixture Experiments

Our results indicate that air space is probably needed for the

storage of a dry mixture of milt and eggs, since sperm storage does require an air space (Truscott et al., 1968). At Big Creek, no air space was provided, and the fertilities after 19 and 30 hours were 42 and 49 percent, compared to 84 and 86 percent at Trask River, where an air space was provided.

SUMMARY

1. The goal of the streamside incubator program is to develop a system, which by simulating conditions in a good natural spawning bed, will produce alevins showing good growth and survival and hatchery fry with characteristics comparable to those of wild fry.

2. Experiments on the effects of water velocity, crowding, and substrate suitability conducted at the OSU Marine Science Center during 1967-68 established design criteria for the construction of the prototype incubator. The incubator was constructed to handle 1.2 million eggs on 382 square feet of crushed rock substrate (about three eggs/cm^2) with water velocity set at 25-50 cm/hr.

3. From sources in Alaska, Washington, and Oregon, about 300,000 pink salmon eggs, 110,000 coho eggs, 100,000 chinook eggs, 6,000 chum eggs, and 10,000 "chumook" (a hybrid with chum females and chinook males) were used to stock the incubator.

4. The problem areas of the prototype were: an unreliable water system, a decreased pump efficiency, siltation, fungus growth on dead eggs, poor circulation in hatchery tanks, and leakages in the tanks. Unusually cold weather did not affect the hatchery operation nor the survival of the salmon.

5. The first year produced approximately 12,000 pink, 28,000 coho, 75,000 chinook, several thousand chum, and 100 chumook. Mortality thus exceeded production. Much of the mortality, however,

was due to catastrophic circumstances, i. e. an intake washout causing a water stoppage of about 14 hours, suffocating about 288,000 pink salmon; a handling error which killed 33,000 newly fertilized coho eggs; an unsuccessful hybridization attempt with about 16,000 coho eggs and chum salmon milt.

6. Less than one percent of the fish in the hatchery exhibited abnormalities or diseases. Alevins reposed quiescently on the gravel substrate and attempted to dig for cover only when disturbed. Emerged fry later schooled and swam in circular patterns within the hatchery tanks.

7. Experiments conducted at Netarts Bay in 1968-69 refined the original design criteria. The results suggested that the best growth and survival are afforded by low initial egg stocking density of about one layer and water velocity from 54 to 220 cm/hr. Relative to the two other substrates tested (crushed rock and screen), a smooth cobble substrate provided cover from light for the developing alevins, and the resulting fry exhibited the greatest fright reaction to disturbance.

8. The following design improvements were suggested to increase the reliability of the hatchery: 1) Remove intake system from the creek to avoid the possibility of it being washed out during a freshet. 2) Provide a filtration system to eliminate silt. 3) Increase water velocity in hatchery tanks to 54 cm/hr or higher. 4) Provide a backflush system to assure good circulation. 5) Elevate eggs off

substrate to facilitate assessment and removal of mortalities. 6) Provide a smooth cobble substrate in which the alevins can hide. 7) Provide an auxiliary power source to back up the electric pump. 8) Eliminate leaks from hatchery tanks. 9) Have a hatcheryman in residence.

9. Research on means of transporting freshly spawned salmon eggs resulted in field tests of a delayed fertilization method whereby dry eggs and milt may be packed separately under cold storage at 6°C and fertilized any time up to 20 hours later without loss of the initial fertility.

10. Delayed fertilization was employed successfully in transplanting an estimated 150,000 pink salmon eggs from Alaska to Oregon in September of 1968, resulting in 92 percent fertility after about 16 hours in storage. Survival of about 200,000 fertilized pink salmon eggs was only about 60 percent after the trip.

11. Two field tests were conducted in Oregon in October - November 1968 to replicate the techniques used in the pink salmon transplant and to investigate other spawning and storage techniques. The results indicated that up to 30 hours, fertilities of 80 percent or higher may be expected from delayed fertilization of coho eggs. Also, the cold storage of a dry mixture of eggs and milt may give comparable fertility to that of delayed fertilization.

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