

Northwest and Alaska Fisheries Center Processed Report*

ESTUARINE AND FRESHWATER CULTURE OF 1974 BROOD COHO SALMON
IN NET PENS AND FLOATING RACEWAYS AT LITTLE PORT WALTER, ALASKA

by

William R. Heard, Roy M. Martin, and Alex C. Wertheimer

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BACKGROUND AND OBJECTIVES

In 1972 the National Marine Fisheries Service (NMFS) began a cooperative research program with the Alaska Department of Fish and Game (ADF&G), Division of Fisheries Rehabilitation Enhancement and Development (FRED) at the Little Port Walter (LPW) field station to test and develop concepts of fry-to-smolt culture of salmonids in estuarine environments. This program grew out of previous experimental work in net-pen culture of salmonids at LPW and elsewhere and was designed to test the biological feasibility of alternative approaches to standard hatchery culture programs.

While some fry-to-smolt stage culture of sockeye salmon (Oncorhynchus nerka) and chinook salmon (O. tshawytscha) has been done at LPW in this program, the principal emphasis has been on coho salmon (O. kisutch). Other relatable studies, including the short-term rearing of pink and chum salmon fry (O. gorbuscha and O. keta) before they are released into the marine environment, have also been conducted at the LPW facility.

Fry-to-smolt stage culture in net pens of species normally going to sea as smolts plus short-term net-pen culture of species normally going to sea as fry, cultured for release and subsequent anadromous adult returns (Ocean Ranching), can be differentiated from net-pen culture of salmonids to produce harvestable size fish in captivity (Ocean Farming). Ocean Farming of salmonids in net pens has received considerable attention in the Puget Sound area (Novtony, 1975), southern British Columbia (Kennedy et al., 1975, 1976a, 1976b), other parts of Canada (Brett, 1974), and elsewhere.

Coho salmon fry from four broods have been cultured over 1-year periods and released at LPW since 1969. Two of these, 1968 and 1969 broods discussed by Heard and Crone (1976), produced adult returns to the LPW area of 12.3 and 16.4% of the numbers of juveniles released.

Fry-to-smolt stage culture of 1971 brood coho salmon (Heard and Martin, 1973; Heard, Martin, and Wertheimer, 1973) involved the first attempt at LPW to raise moderately large numbers of fry in small-mesh net pens. Epizootics, caused principally by the marine bacterium Vibrio anguillarum, killed over 90% of the initial 285,000 coho salmon fry during the summer of 1972. Only 10,800 age 1 fish from the 1971 brood were released at LPW in 1973. The total number released comprised several individual lots marked with excised left ventral fins and liquid nitrogen brands. Due to poor growth and complications associated with disease outbreaks, almost one-third of those released were still in parr stage.

The overall age 3₂ adult return of 1971 brood coho salmon to LPW in 1974 was 3.2% of the number of juveniles released. Return rates for individual lots ranged from 0 to 9.9%.

The culture of 1972 brood coho salmon at LPW was the second attempt to raise substantial numbers of fry to smolt stage (Heard, Martin, and Wertheimer, 1974, 1975). Vibriosis epizootics were also encountered with culture of these fish, both during the primary growth Phase 1 period (summer 1973) and during the overwinter Phase 2 period (1973-1974). Disease losses were not as severe with 1972 brood juveniles as they were with 1971 brood fish. The improvement in survival, we believe, was due to experiences gained in early-stage fry culture in net pens and with changes in methodology. From the initial 425,000 1972 brood fry, 280,000 or 65.8% survived through the Phase 1 period, 186,000 or 43.8% (of the initial number) survived through the Phase 2 period. By the end of the Phase 3 period (spring 1974) 173,000 smolts were released at LPW for an overall fry-to-smolt survival of 40.7%. Age 3₂ adult returns from these smolts in 1975 amounted to 8.7% of the number released. Survival from eight individually-marked lots ranged from 1.1 to 8.9% and averaged 8.0%.

In 1973 we began experimental work at LPW with the use of single-pass flowthrough floating raceways made from nylon-reinforced impervious plastic fabric. These were initially used as a complimentary tool for early-stage fry culture in net pens. Subsequent uses of the floating raceways have led to a variety of modifications in the original fry-to-smolt stage strategy that utilized only net pens. These modifications now include culture in fresh water and controlled intermediate salinities in various temporal combinations throughout the fry-to-smolt stage culture period. Design, construction, and operational details of the floating raceways are discussed by Heard and Martin (1976).

Estuarine net pens depend on tidal action to maintain the water flow necessary for a suitable culture environment while floating raceways require a controlled flowthrough water source similar to those used with concrete raceways and other standard rearing containers. A significant portion of the activity at LPW in 1974 was associated with developing a suitable year-round water system for use with the floating raceways. This involved installation of an 8-inch-diameter polyvinyl chloride (PVC) waterline from just above Sashin Creek Falls to a point beneath the fish culture float in the inner part of LPW Bay.

The primary objective of this report is to describe the freshwater and estuarine culture of 1974 brood coho salmon in floating raceways and net pens. Emphasis is placed on: (1) documenting the numbers, survival and growth of fish in various populations during each of the three culture phases (Phase 1, 2, and 3); and (2) describing the culture treatments, rationale for the treatments and the numbers of marked and unmarked smolts released from each of these treatments.

Secondary objectives of this report are to: (1) briefly describe the rationale, planning, and installation of the 8-inch PVC pipeline in 1974 that makes the use of floating raceways at LPW possible; and (2) consider the sources of 1974 brood coho salmon eggs collected at LPW, the incubation schemes used with those eggs, and description and results of some preliminary evaluations of incubation tests relative to initial fry quality.

WATER SYSTEM DEVELOPMENT

The initial planning for developing a water system capable of providing 1 to 2 cfs (450-900 gpm) flow for experimental fish culture at LPW began during the summer of 1973. Organization of the project, including procurement of the required permits and necessary materials, continued throughout the fall, winter, and early spring period of 1973-1974. The proposed project centered around installation of about 5850 feet of 8-inch PVC pipe in a remote area consisting of mixed spruce forest-muskeg meadow terrain without heavy construction equipment. Installing the waterline without heavy equipment was necessary to avoid undue environmental disruption along the lower part of Sashin Creek. A document describing the planning, permits, and environmental assessments of this project is on file at the Auke Bay Laboratory (Anonymous, 1974).

The basic plan for installing the 8-inch waterline called for roughly paralleling the route of two smaller waterlines. Both of these were 2-inch-diameter polyethylene pipes that originated at Sashin Falls and traversed along the south bank of the stream and across the inner LPW Bay to the vicinity of the station headquarters. These 2-inch waterlines were installed in 1969 to provide domestic water and a small amount of incubation water. Collectively they provide about a 50 gpm flow. The general layout of the Port Walter vicinity showing the approximate location of the waterlines and other local areas mentioned in this report is shown in Figure 1.

The 8-inch waterline was a NMFS-FRED Division project. A special cooperative agreement was developed regarding the cost, utilization, and ultimate ownership of the system. One of the initial reasons leading to this project was the need to raise substantial numbers of coho salmon fry for 1 to 2 months in fresh water before planting them in a nearby lake. Most of the cost of materials and much of the labor cost for the new waterline was paid from lake study funds, a separate cooperative program for research on underutilized lakes near LPW.

CONSTRUCTION

Construction of the 8-inch waterline began in May, 1974 as soon as snow cover had melted sufficiently to permit access to the area. Materials and supplies were transported to LPW from Juneau via the NMFS vessel Murre II. The pipe itself, mostly in 20-foot lengths, was

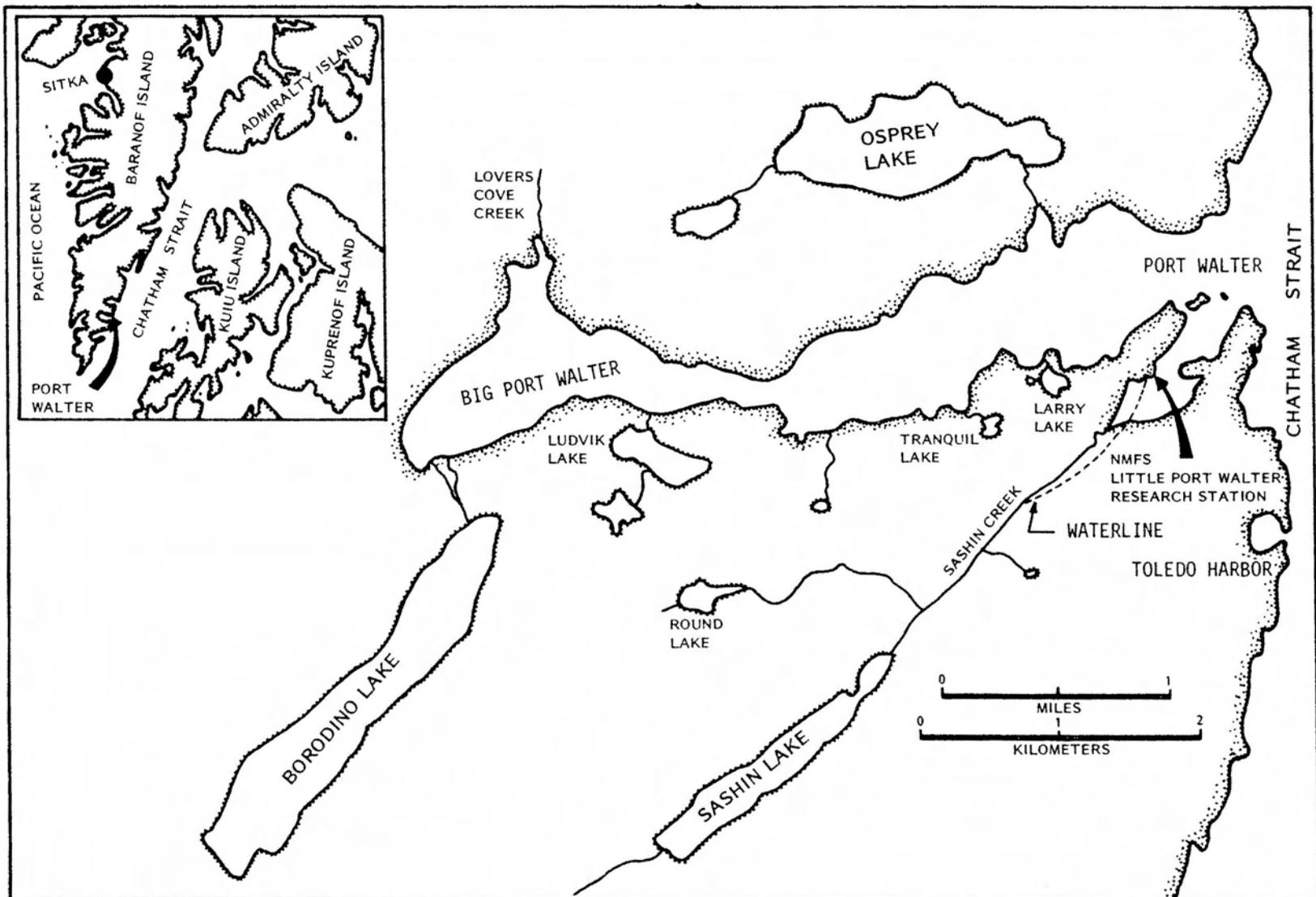


Figure 1.--Port Walter Drainage Area, South Baranof Island, Alaska, showing the location of NMFS Little Port Walter Biological Field Station and adjacent areas involved in fishery research activities.



Figure 2.--Helicopter transport of 20-foot lengths of 8-inch-diameter PVC waterpipe from intertidal portion of Little Port Walter Bay to waterline route along Sashin Creek.



Figure 3.--Installation of 8-inch-diameter PVC waterline in spruce hemlock forest area along Sashin Creek. Two smaller, 2-inch-diameter, polyethylene waterlines (black pipe in photo) paralleled part of the route of the larger pipe.



Figure 4.--Muskeg meadow crossing of 8-inch-diameter PVC waterline. Note the O-ring coupler used to join lengths of pipe together.

airlifted by helicopter from the intertidal portion of LPW Bay (Figure 2) and deposited along the route.

Installation required burying the waterline approximately 6 inches in forest, muskeg, and intertidal areas. Excavation for burial of the pipe was done by hand or by "trenching" with explosives. Usually a single three-man crew or two two-man crews worked on installation. Occasionally larger crews were used. The waterline route traversed approximately 4250 feet of forest or mixed forest-muskeg area (Figures 3 and 4), 700 feet of beach and intertidal area, and 900 feet across the floor of LPW Bay.

TESTING

Construction of the waterline, from the well-screen intake structure in Sashin Creek to the lower part of the intertidal area was completed by mid-September, 1974. An inline valve and tee, near Sashin Creek weir at the stream mouth, permitted flow through the upland portion of the waterline without flow through the intertidal and submerged parts of the system. Initial test flows at Sashin Creek weir approached the design flows of about 2 cfs.

The subtidal portion of the waterline was completed during October and November, 1974. Divers were required for this part of the project. Considerable difficulty with the O-ring couplers that joined lengths of pipe together underwater was experienced when flow was directed through the submerged part of the waterline. Also, extreme care was required when directing waterflow into this part of the system to avoid forcing a column of air underwater (inside the pipe) and causing the pipe to float to the surface. Details of terminal delivery of water flow from the bay floor to the bay surface are covered in a discussion on floating raceways (Heard and Martin, 1976).

COLLECTION OF ADULTS AND EGGS

Adult coho salmon returning to the LPW area in 1974 were collected at Sashin Creek, Tranquil Creek, and Ludvik Creek. Tranquil and Ludvik Creeks are the outlet streams from Tranquil and Ludvik Lakes (Figure 1). Adults returning to Sashin Creek originated primarily from wild smolts that emigrated from the stream in 1973, and cultured smolts from 1971 brood progeny, raised from fry-to-smolt stage in estuarine pens and released in the LPW area in 1973. Adults returning to Tranquil Creek and Ludvik Creek originated primarily from age 1 "semi-wild" smolts emigrating from Tranquil and Ludvik Lakes in 1973. Both lakes were planted with 1971 brood coho salmon fry in June, 1972. Neither lake contained any fish when they were stocked with fry.

The total number of adult coho salmon returning to each of the three areas and the disposition of those adults is as follows: Sashin

Creek, 690 adults (367 into stream for natural spawning, 323 into holding pond for ripening and artificial spawning); Tranquil Creek, 133 adults (all into holding net off stream mouth for ripening and artificial spawning) and Ludvik Creek, 531 adults (all into net pens off stream mouth for ripening and artificial spawning). There is no natural spawning in Tranquil or Ludvik Creeks because of barrier falls near tidewater.

Although adult coho salmon returning to the LPW area in 1974 came from four groups of smolts at three locations, all originated from Sashin Creek stock. All previous lake-stocking and estuarine culture studies with coho salmon in the LPW area have used Sashin Creek fish or progeny from fish that ultimately originated in Sashin Creek. Many of the adult coho salmon returning to Sashin, Tranquil, and Ludvik Creeks in 1974 could be identified with the originating source of smolts from specific marks.

Approximately 1.2 million coho salmon eggs were artificially spawned at LPW in 1974. Of this number, about 630,000 eggs were retained at LPW for incubation and 570,000 were shipped green (immediately after spawn-take with gametes separate) to the ADF&G Crystal Lake Hatchery near Petersburg. Spawning began in early October and terminated in early November. The eggs retained at LPW were spawned mostly during the first part of this period.

Because some of the adults returning in 1974 bore marks of specific groups of smolts, certain lots of eggs from known parents were kept separate from others. Eggs identified as "lake stock" came from parents known to have originated from smolts emigrating from Tranquil or Ludvik Lake while eggs identified as "EPR stock" came from parents known to have originated from estuarine-pen-raised smolts. The primary purpose of keeping these egg lots separate was to investigate the possibility that progeny of survivors from severe vibriosis epizootics develop a natural immunity against the disease. EPR stock adults returning with LV marks in 1974 had survived the vibriosis outbreak during estuarine pen culture in 1973. Lake stock adults returning with RV or Dorsal marks in 1974 had grown from fry-to-smolt stage in a lake environment and had not experienced a vibriosis epizootic during that period.

EGG INCUBATION AND FRY SOURCES

All 1974 brood coho salmon eggs retained at LPW were incubated from green egg to emergent or swim-up fry stage in a small, experimental wet laboratory facility located in the main warehouse. Most eggs were seeded "green" at high densities in Heath incubator trays until they reached early eyed stage. After reaching eyed stage, dead eggs were removed and the live eyed eggs were reseeded in one of three primary incubation modes: (1) Heath incubator trays, (2) deep matrix gravel incubators with upwelling flow, and (3) prototype Auke Bay Incubator (stacked trays with astroturf). The eggs were treated weekly with malachite green to control fungus during the green egg to advanced eyed egg period.

One group of 87,500 eggs was seeded green in a seven-tray Auke Bay Incubator unit to evaluate the removable egg trays and special alevin drop-through feature that this incubator provides (Salter, 1975). About 50% of these eggs died before hatching and only 31,200 fry ultimately survived from this group of eggs.

Few eggs died in Heath trays during the green to eyed egg period. Heath trays were seeded at 4600 eggs per tray (1.2 liters) for incubation during the eyed egg to fry state. During the eyed egg to fry period, an airlock in one 16-tray Heath incubator unit caused a 97% loss of alevins. Two trays in one Heath incubation unit were lined with astroturf to compare fry quality from rugose and smooth surfaces.

The numbers of green and eyed 1974 brood coho salmon eggs seeded in various incubation schemes at LPW, the numbers of fry, survival values, and average weight of fry produced from each are summarized in Table 1. The overall survival for various incubation stages were: (1) from green egg to eyed egg stage, 92.2%; (2) from eyed egg to fry stage, 84.1% and (3) from green egg to fry stage, 77.5%.

FRY QUALITY

The comparative mean weights of fry from various incubators in Table 1 were derived from en masse weighings of 500- to 2000-g samples of fry. To do this, a weighing container was tared with about 250 g of water on a tri-beam gram balance. The fry (to be weighed) were drained to an attempted standard end point in a perforated polypropylene "strainer box" then poured into the tared container and weighed. The number of fry in the sample are counted to determine mean weight. Pooled samples from Heath and gravel incubators indicated that fry from the gravel incubators weighed about 10% more than those from Heath incubators (Table 1).

To more accurately measure differences in the relative size of coho salmon fry produced from the different incubators, 50 to 60 fish samples were collected from the incubators and preserved in 5% formalin for individual measurements of length and weight. After allowing preserved fry to stand 6 weeks, individual lengths were measured to the nearest millimeter and wet weights to the nearest milligram. This procedure permits calculation of an index to developmental stage or K_D value (Bams, 1970) to account for relative yolk material remaining when the sample is taken. Detailed studies on incubation evaluation of pink salmon fry (Bailey and Taylor, 1974; Bailey, Pella, and Taylor, 1975) have found this index of development useful when considering fry quality relative to timing of emergence, and emigration of unfed fry.

The fork lengths, weights, and K_D values determined for four types of incubation (Table 2) indicate that regular Heath incubator fry tend to be more developed (smaller K_D values indicate less yolk reserve) than

Table 1. Summary of 1974 brood coho salmon eggs incubated at Little Port Walter.

Type of Incubation	Numbers of green eggs seeded	Numbers of eyed eggs produced or seeded	Numbers of fry produced	Survival (percent)	Mean weight of fry (mg) ^{1/}
Heath trays eyeing only	538,000	531,500	--	98.8	--
Heath unit 1 16-trays		73,600	2,300	3.1	263
Heath unit 2 16-trays		73,600	73,600	100.0	263
Heath unit 3 15-trays		69,000	69,000	100.0	263
Heath unit 4 15-trays		69,000	69,000	100.0	263
Gravel unit 1		75,900	75,700	99.7	290
Gravel unit 2		82,800	73,400	88.6	290
Gravel unit 3		75,900	77,300	101.8	290
Auke Bay Astroturf 1-tray		12,500	13,000	104.0	263
Auke Bay astroturf 7-trays	87,500	43,750 (to hatching)	31,200	50.0 to 35.7 to fry	288
Overall survivals:					
green egg to eyed egg	625,000	576,050		92.2	
eyed egg to fry		576,050	484,500	84.1	
green egg to fry	625,500		484,500	77.5	

^{1/} Based on 500 to 2000 g. samples of fry; values determined for fry pooled from individual Heath and gravel incubators.

fry incubated in gravel or astroturf substrates. A dramatic increase in mean weight and yolk reserve occurred over normal Heath fry when a single layer of astroturf covered the bottom screen of the Heath basket (Table 2).

We made comparisons of mean fry weights determined from an en masse weighing of live fish and individual weights of preserved fish collected from a common population at the same time. The comparison was made with fry collected from Heath trays modified with astroturf on April 20, 1975. Two groups of preserved fry (weighed after 6 weeks preservation in 5% formalin) averaged 305 and 308 mg per fry. The 1374.1-g sample of live fry contained 4496 fish for a 306-mg-per-fry average, closely paralleling the preserved fry samples.

En masse weighings of live fry (and larger fish) are biased upward (from true weight) because of cumulative moisture content and difficulty in arriving at a standard end point when draining the sample without unduly stressing the fish. Large samples tend to standardize cumulative moisture bias but likely add to the stress problem (which may change relative to different sizes of fish). With live fry, we have usually achieved reproducible results by avoiding fry sample sizes < 500 g. Moisture content bias from weighing individual fish is presumably minimized by blotting; however, formalin-preserved fish increase in weight over live weight (Parker, 1963), so there are compensating biases in the two procedures. On the basis of this comparison, it appears that en masse weighings of live fry did provide suitable estimates of mean fry weight.

TIMING OF EMERGENCE

The timing of coho salmon fry emergence from substrate incubators and the "ponding" or removal of fry from Heath trays is an important event that influences subsequent culture activity. Although fry voluntarily emerging from a gravel incubator are likely to be larger than fry from Heath trays, emergence may cover a 5- to 6-week period. This tends to spread out the initial feeding period, and the population may develop a wide length frequency distribution since early emerging fry are fed several weeks longer than late emerging fry.

Fry began emerging from the three gravel incubators in late March, 1975. However, retainer screens over the outlet drains prevented early emerging fry from emigrating from the units, a step that eliminated the need for separate holding facilities for the early fry. This caused a substantial build-up of fry in the water column above the incubation gravel before the screens were removed on April 19. The pattern of fry emergence from the three incubators (Figure 5) indicated that approximately 26, 42, and 46% of the total emergence had occurred by April 20 when daily counts were started of emigrant fry from each incubator.

Table 2. Lengths, weights, and stage of development for 1974 brood coho salmon fry (unfed) from four types of incubators at Little Port Walter.

Type of Incubation and date of samples	Number of fry	Mean length (mm)	Mean Weight (mg)	Mean Developmental Index (K_D)
<u>Upwelling Gravel</u>				
April 23, 1975	60	30.4	296.3	2.191
April 23, 1975	58	30.9	296.3	2.158
<u>Normal Heath Tray</u>				
April 20, 1975	63	31.5	287.7	2.092
April 26, 1975	62	31.8	279.7	2.053
<u>Modified Heath Tray with Astroturf</u>				
April 20, 1975	55	31.7	305.4	2.121
April 20, 1975	61	31.6	308.2	2.134
<u>Auke Bay Tray with Astroturf</u>				
May 1, 1975	59	30.4	295.0	2.187
May 1, 1975	61	30.8	278.4	2.118

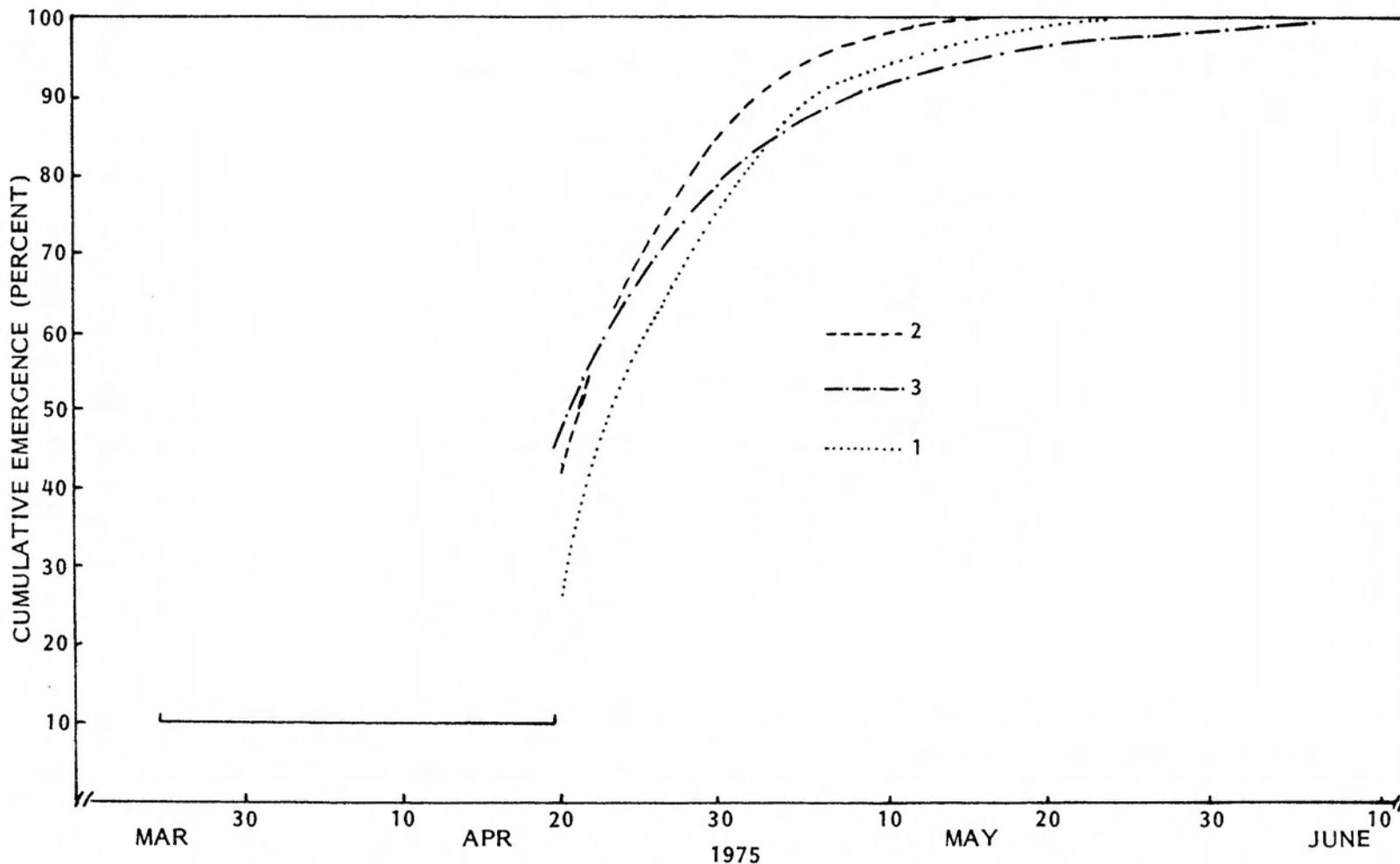


Figure 5.--Cumulative percent emergence of coho salmon fry from three gravel incubators at Little Port Walter during 1975. The horizontal line includes the period when emergence was occurring but was not enumerated daily because retainer screen kept fry in the water column above the natal gravel. Cumulative early-emerging fry were collectively enumerated from each incubator on April 19.

Fry originating from Heath trays are usually ponded for initial feeding when most of the yolk material is absorbed. This requires deciding when some mid-point is reached between fry with remaining yolk and others that have the yolk completely absorbed or "buttoned up." We periodically tested samples of alevins from Heath trays by placing them overnight in a small open container of water to determine how many became swim-up fry. (A working definition of alevins emerging into fry includes the initial filling of swim bladders by gulping air bubbles, achieving neutral buoyancy, and swimming horizontally in the water column). When about 25% of the Heath incubator alevins reached the swim-up fry stage we removed the trays and placed all alevins and fry into their initial rearing containers. The remaining alevins generally became swim-up fry in 3 to 4 days. We ponded all Heath incubator fry on April 19 and 20.

Fry originating from the one Auke Bay incubator tray seeded with eyed eggs were ponded on May 2, 1975, while fry emerging from the seven-tray Auke Bay Incubator unit were allowed to emigrate from the trays between April 27 and June 13. A special feature of the Auke Bay Incubator is the ability to permit voluntary emergence or to retain the fry in individual trays for subsequent ponding by the operator.

FRY-TO-SMOLT STAGE REARING

Earlier rearing of juvenile salmonids at LPW has led to defining culture activities in terms of three seasonal periods or phases (Heard, Martin, and Wertheimer, 1975). Phase 1 begins with swim-up fry in spring and includes the primary growth period throughout the first summer of life (usually 3 to 4 months). Phase 2 begins in fall when water temperatures are declining and includes the overwinter and early spring period when growth is minimal (usually 5 to 6 months). Phase 3 is the terminal spring period when temperatures and photoperiod are rapidly increasing and significant growth is possible before the age 1 fish are released (usually 2 or 3 months).

The initial releases of estuarine-cultured coho salmon smolts at LPW were made at the end of the Phase 3 period (around June 1) to coincide with peak emigration of wild smolts from Sashin Creek. Subsequent releases of marked smolts from LPW at other time periods and the adult returns from these releases suggest that reasonably good ocean survivals can be achieved from releases made over a broadened time interval. One of the primary objectives of 1974 brood coho salmon culture at LPW is to continue investigation of the effects of time and size at release on ocean survival along with the various treatments of estuarine and freshwater culture in raceways and net pens.

PHASE 1 CULTURE (FIRST HALF, APRIL 19 TO JULY 17)

A wide variety of holding containers were used for initial ponding and feeding of coho salmon fry from various incubators. This start-up period began April 19 and was terminated June 13, the last date new fry were added to any population from incubators. The initial holding containers used included: (1) a number of 4- by 8- by 4-foot by 1/8-inch-mesh net pens suspended inside rectangular-shaped floating raceways; (2) a plastic-lined circular swimming pool 20-foot diameter by 4-foot depth located behind the main warehouse (started May 20); (3) two floating horizontal raceways, each 62- by 14- by 7-foot (started May 6 and June 19); and (4) two floating conical-shaped vertical raceways each 13- by 6- by 10-foot (started May 22 and June 10).

The rectangular-shaped floating raceways did not satisfactorily hold their design shape in the estuary (partly because they were filled with fresh water) and their use was discontinued after 2 to 3 weeks as other holding containers became available. Both the horizontal raceways (Figure 6) and vertical raceways (Figure 7) have since become useful fish culture tools at LPW.

The use of the many temporary holding containers during the first part of the Phase 1 period was the result of: (1) initial testing of new floating raceway prototypes; (2) an initial need to maintain groups of fry separate originating from lake stock and EPR stock parents; and (3) the need to raise a substantial number of fry for 1 to 2 months before they were planted in Osprey Lake. All fry culture during this period occurred in fresh water. As the above steps were completed, the initial fry groups were recombined into more permanent populations.

Growth and Survival

The initial feeding and growth of fry on Oregon Moist Pellet diet (starter mash and 1/32-inch pellets) was slow due to low water temperatures (2 to 3°C) in late April, May, and early June. Growth rates of different groups of fry during this period were all less than 1.0% of body weight per day. As water temperatures in mid and late June increased to 6 to 8°C, growth rates quickly accelerated, approaching or exceeding 3.0% per day.

Overall survival during this period, based on the estimated 484,500 fry produced from various incubators (Table 1) and the estimated 450,500 fry in mid-July, was 92.9%. No disease losses occurred and the principal causes of fry losses were: (1) malformed and pin-head fish removed from initial fry groups, and (2) mink (Mustela vison) predation. All of the initial rearing units were covered with gillnet webbing but the meshes proved to be too large and mink were seen readily entering and leaving several units. A combination of a 2-foot-high, small-mesh wire fence around the units and a small-mesh webbing cover over them was subsequently

developed to keep out mink. Three problem animals were livetrapped and removed from the area.

Planting Fry in Osprey Lake

On July 15, 276,000 coho salmon fry were planted in Osprey Lake from the initial population started in a horizontal raceway on May 6. The fry averaged 359 fish per pound (1.26 g/fish). These fry originated primarily from parent fish returning to Ludvik Lake and Tranquil Lake the previous fall.

Fry were removed from the raceway by crowding them to one end with a small-mesh seine and lift-net (Figure 8). After determining the average weight of fish in the population (similar to the en masse weighing procedure described earlier but with larger scales and tared container), the required pounds of fry (768.9 pounds) were weighed in small lots (Figure 9), transported into a hauling tank inside a De Havlin Beaver floatplane (Figure 10) and flown to Osprey Lake. Eight round-trips with the floatplane between LPW and Osprey Lake were necessary to complete the fry plant. Field activities associated with the lake-stocking studies near LPW in 1975 are reviewed by Crone (1976).

PHASE 1 CULTURE (SECOND HALF, JULY 16 TO OCTOBER 22)

After completion of the fry plant in Osprey Lake, all remaining 1974 brood juvenile coho salmon at LPW were arranged initially in three populations: two in conical raceways (lake stock and EPR stock progeny) and one in the horizontal raceway started on June 19 (mixture of lake and EPR stock progeny). These three groups totaled an estimated 174,500 fish and became the basis for continued intensive culture in floating raceways and net pens. A variety of culture treatments originated from these three populations.

Populations VR-1 and VR-2

Two populations were established to investigate: (1) the possible occurrence of natural immunity to the marine bacterium Vibrio anguillarum in progeny of vibriosis epizootic survivors, and (2) the effects of extended culture (6 to 9 months) at a controlled, intermediate salinity on ocean survival of marked groups of released smolts.

On July 17 two lots of 10,000 coho salmon fry were hand-counted to establish populations VR-1 (control=lake stock) and VR-2 (treatment test=EPR stock). The populations would be cultured in the identical conical-shaped vertical raceways (VR's) at intermediate salinities at least through the Phase 2 (overwinter) culture period. If vibriosis outbreaks developed (as they had during 1972 and 1973 when water temperatures in late July and August reached seasonal highs) and if any



Figure 6.--Floating horizontal raceways used at Little Port Walter in 1975 and 1976.

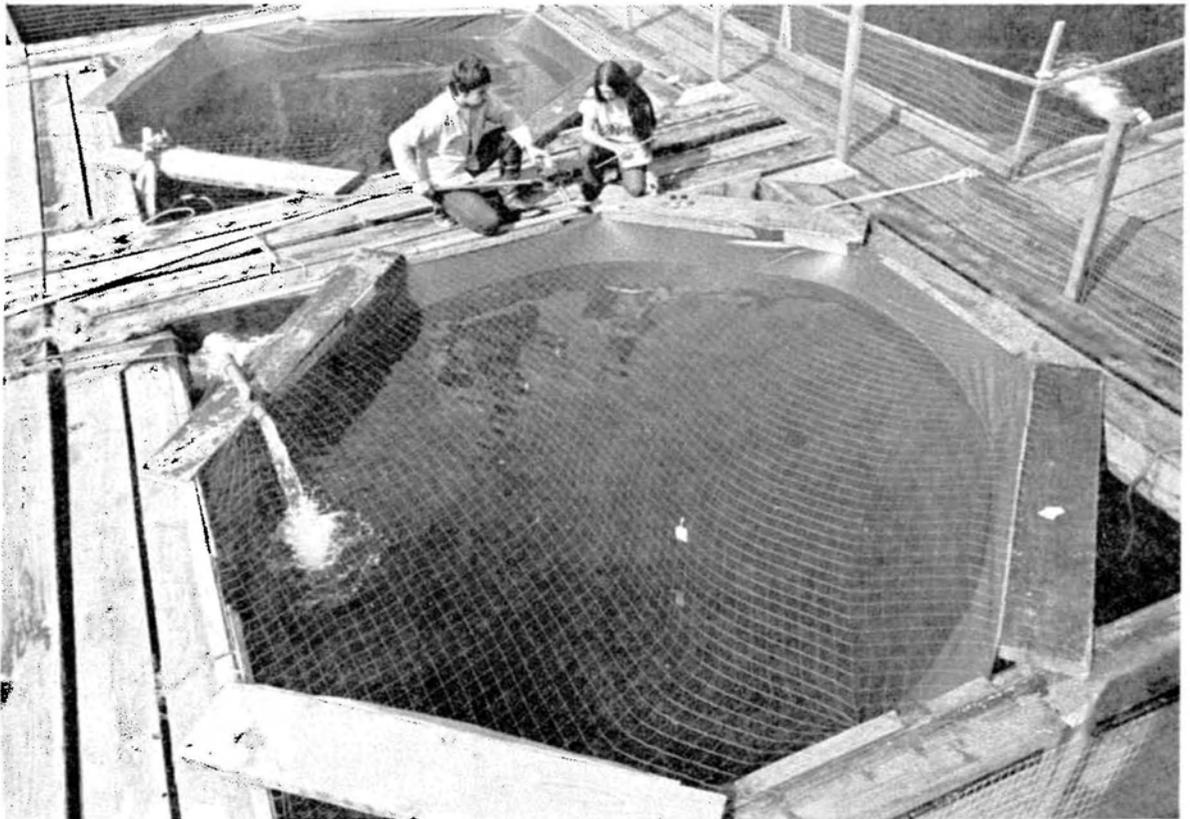


Figure 7.--Floating vertical raceways (conical-shaped) used at Little Port Walter in 1975 and 1976.



Figure 8.--Coho salmon fry crowded to one end of floating horizontal raceway with seine and lift net. Fry are being weighed before transport by floatplane to Osprey Lake.



Figure 9.--En masse weighing of coho salmon fry in a tare-balanced plastic bucket.



Figure 10.--Loading coho salmon fry into special hauling container in a De Havlin Beaver floatplane at Little Port Walter, July 15, 1975, for transport to nearby Osprey Lake.

natural Vibrio immunity in progeny of epizootic survivors did exist, quantification of the immunity should be measured by differential mortality in VR-1 and VR-2 populations.

Relatively constant intermediate saline water in the single-pass plumbing system used for these populations was achieved by using a simple venturi device to inject seawater into the freshwater flow (Heard and Salter, 1976). Water flow into these populations was changed from fresh water to 7 ‰ salinity on July 22. This salinity was readily compatible with the 1.2- to 1.3-g fry and it made exposure of fry to Vibrio possible. Throughout the 6 to 9 months duration of VR-1 and VR-2 (and subsequently VR-3) populations, salinities generally averaged about 8 ‰.

Operational volumes of the vertical raceways were about 665 ft³ (~5000 gallons). Initial flow rates in July and August were maintained around 30 gpm. This provided for an exchange rate or number of changes per hour (R) of 0.4 (Westers, 1970). In September and October, as total poundage increased, flows were maintained around 65 gpm (R=0.8).

Populations HR-1 and HR-2

The numbers of coho salmon fry in the horizontal raceway started on June 19 were periodically increased by combining various fry groups, increasing from an initial 59,000 to 154,500 by July 17. During this 28-day period fry in the raceway grew from 38.1 mm and 0.53 g to 47.9 mm and 1.25 g, for a growth rate of 3.0% per day. These growth rates are approximate due to the cumulative addition of different fish into the population. No additional fry were added to this population (designated HR-0, Table 3) between July 17 and August 13 when it was divided (by weight) into two equal groups to form new horizontal raceway populations HR-1 and HR-2.

The HR-1 and HR-2 populations (plus all earlier start-up fry groups leading to these populations) were cultured in fresh water throughout all of the Phase 1 and Phase 2 periods. The primary purposes of these populations were: (1) continued evaluation of the horizontal raceways, (2) freshwater fry-to-smolt stage culture, and (3) a standard source of freshwater-cultured juvenile coho salmon for developing other population treatments.

Operational volumes of the horizontal raceways were about 3800 ft³ (~28,000 gallons). Flow rates were maintained around 160 gpm (R=0.3) during most of the Phase 1 period.

Populations NP-1 and NP-2: Age 0 Fall Releases

At the end of the Phase 1 period (late October) three new populations of 10,000 fish each were developed from the HR-1 and HR-2

populations. On October 20 and 21 three 5000-fish lots were removed from each horizontal raceway and combined to form the new populations. One group of 10,000 fish was marked with an excised adipose fin and tagged with coded wire tags (ad+cwt, data 1/11) and released on October 22 to test age 0 fall releases at the end of the primary growing season. The other two groups of 10,000 fish (both hand-counted) were placed in two net pens to become NP-1 and NP-2 populations for overwinter culture in the estuary. Both net pens were 12- by 25- by 12-feet by 1/4-inch-mesh webbing. Juvenile coho salmon in the three new populations averaged 8.4 g and 91.0 mm.

Age 0 fall releases of coho salmon "smolts" in Alaska is frequently discussed and it has certain appeal because it would forego expensive overwinter culture (normally a difficult time at best) required to release the same fish at about the same size the following spring. At the present time, however, there is no demonstrated biological basis for expecting adult returns from releases of age 0 fish made during the fall.

Overwinter culture of NP-1 and NP-2 populations (at 29 ‰ to 31 ‰ seawater salinities) is consistent with previous estuarine fry-to-smolt culture work in LPW Bay. Survivors in NP-1 were scheduled for differential marking and released at the end of Phase 2 culture (early April) while for those in NP-3 at the end of Phase 3 (late May). A fourth new population of about 10,000 fish was also scheduled for marking and release from the horizontal raceways at the end of the Phase 2 period after overwinter culture in fresh water. The experimental design of these four populations was arranged so that adult return data could be correlated to the original prewinter groups of 10,000 fish.

Growth and Survival

The VR and HR populations were sampled periodically from July through October to determine mean weight using 5- to 20-pound subsamples of fish from the populations. Mean length was usually determined from measurements of 100 to 200 fish from these subsamples. Estimating population size (total pounds and numbers of fish) was determined less frequently to avoid undue handling and stress. In this later procedure, biomass of the population is determined by weighing all the fish in small lots (see Figure 9) to determine total weight then dividing by the mean fish weight to estimate total number.

Growth rates that remained above 3.0% of body weight per day during August, dropped to less than half this rate (1.2 to 1.4%) in September and further declined in October to 0.4 to 0.7%. Changes in rate of growth were likely related not only to declining temperatures that began in September (Figure 11), but also to declining photoperiods plus possible decreases in overall growth potential resulting from increased size of the fish (Brett et al., 1969). Growth rates for individual populations during this period are reviewed in Table 3.

Figure 11.--Seasonal temperature patterns encountered in various culture modes at Little Port Walter during fry-to-smolt rearing of 1974 brood coho salmon. Ambient conditions are represented by fresh water (recorded from freshwater raceways) and seawater (recorded from 2-m depth in estuary), while modified conditions are represented by intermediate salinity raceways. All groups smoothed by a 10-day moving average.

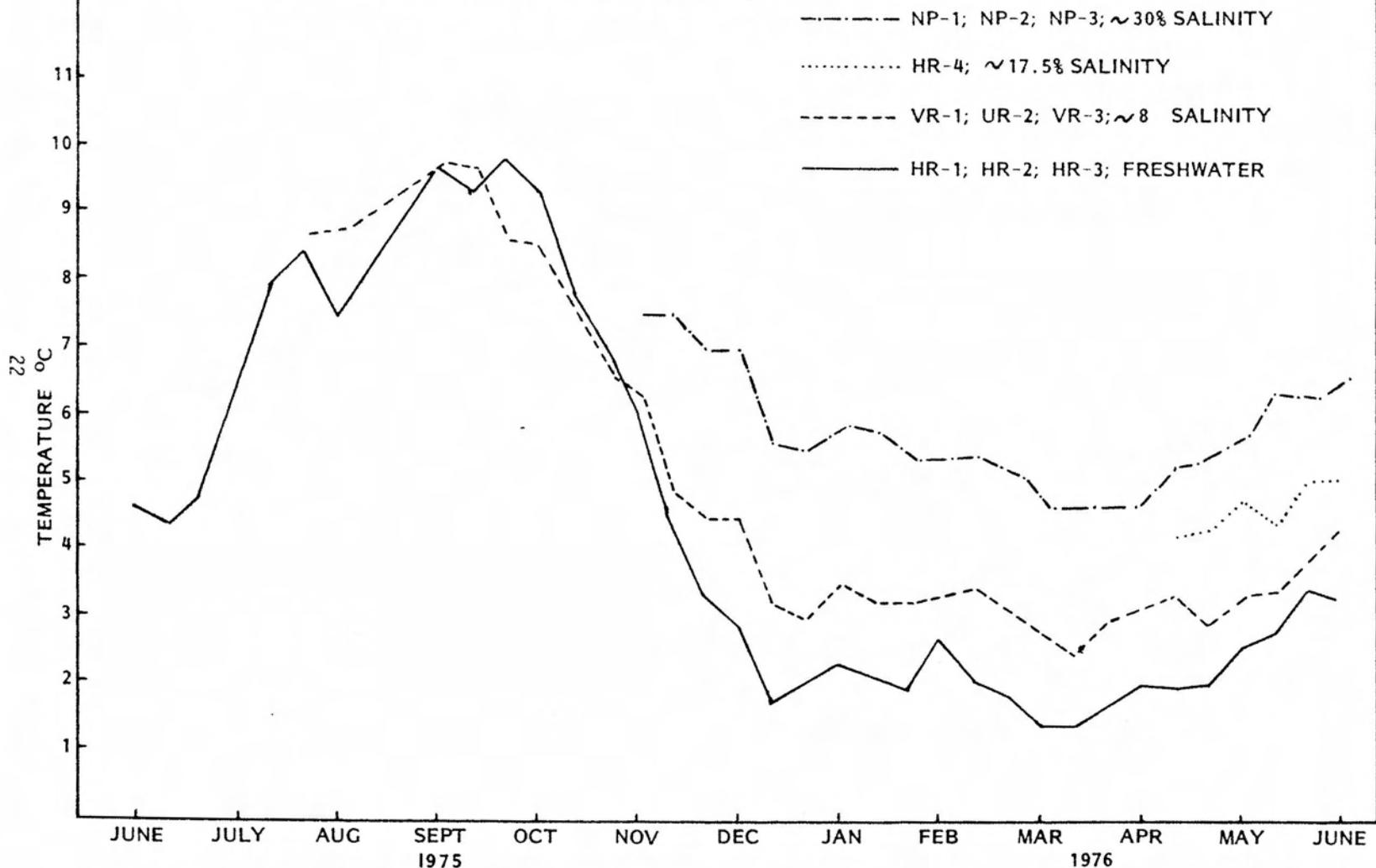


Table 3. Comparisons of lengths, weights, numbers of fish, growth, and survival rates in five cultured populations of 1974 brood coho salmon at LPW between July 17 and October 22, 1975.

Population	Sample date	Mean fork length (mm)	Mean weight		Population size		Rate of growth ^{1/} (percent)	Survival
			fish per pound	grams per fish	pounds	number (thousands)		
HR-0	July 17	47.9	362.0	1.25	426.8	154.5	3.1	101.7
	Aug. 13 ^{2/}	60.6	161.5	2.81	973.7	157.2		
HR-1	Aug. 13	60.6	161.5	2.81	487.0	78.6	0.6	98.7
	Aug. 31	72.2	92.4	4.91	-	-		
	Oct. 1	86.6	61.0	7.44	-	-		
	Oct. 20	91.2	53.6	8.47	1440.0	77.6		
HR-2	Aug. 13	60.6	161.5	2.81	486.7	78.6	0.6	99.5
	Aug. 31	70.9	93.4	4.86	-	-		
	Oct. 1	86.2	60.5	7.50	-	-		
	Oct. 21	90.7	53.9	8.42	1451.0	78.2		
VR-1	July 17	47.9	374.3	1.21	26.7	10.0	0.4	95.0
	Aug. 31	72.4	91.5	4.96	-	-		
	Oct. 1	84.2	61.0	7.44	-	-		
	Oct. 22	87.5	56.5	8.04	168.4	9.5		
VR-2	July 17	49.2	350.1	1.29	28.6	10.0	0.7	96.0
	Aug. 31	71.7	94.7	4.79	-	-		
	Oct. 1	81.5	66.3	6.85	-	-		
	Oct. 22	87.3	57.3	7.96	167.6	9.6		

^{1/}Percent increase in body weight per day; derived from the equation: $\frac{1}{t} \ln \left(\frac{W_1}{W_0} \right)$; where W_1 is the mean weight at the end of a time period, W_0 is the mean weight at the beginning of the time period and t is the number of days in the time period.

^{2/}HR-0 was weighed into two equal groups to form HR-1 and HR-2 on August 13.

Overall survival, based on an estimated 174,500 fish at the beginning and 174,900 at the end of the period, exceeds 100%. The principal reason for this anomaly was a 2.7-thousand-fish increase in estimated numbers in population HR-0 between July 17 and August 13 when it was divided into HR-1 and HR-2 (Table 3). Past experiences during fry-to-smolt culture of coho salmon at LPW with biomass estimates of population numbers (total weight ÷ mean fish weight = estimated population) have suggested that the procedure, even under carefully standardized conditions, has a 3 to 5% margin of error.

Parasites and Disease: Occurrence and Treatment

A few fish began dying in the vertical raceways in early August. Samples of distressed and apparently healthy fish from these populations were sent to the FRED Fish Pathology Laboratory in Anchorage on August 8 for analysis. Bacterial isolates from this analysis included Aeromonas and Pseudomonas but no Vibrio. The loss of fish in these intermediate saline populations never increased above 4 or 5 fish per day and even this rate declined in September and October. The 4 to 5% loss of fish in these populations between July and October (Table 3) was attributed to a low level bacterial infection.

Heavy "flashing" and "scraping" behavior began occurring in population HR-0 in early August, suggesting a possible external protozoan infection. A sample of 10 healthy and 10 sick fish sent to the Pathology Laboratory for examination on August 4 failed to reveal organisms that might have been causing the flashing behavior. A 1:13,000 flow-through formalin treatment given to this population on August 5 essentially eliminated the flashing within 4 h of the treatment. The flow-through treatment was preceded with three 1-h static bioassay tests in 1:4000 formalin baths to insure that it would not jeopardize the population (Wood, 1974). During the treatment, 2.3 gallons of formalin was applied (at a 0.5% solution) over the raceway during a 4-minute interval. Turnover rate of the raceway with 160 gpm flow was about 3 h.

Two 10-day drug therapy treatments of Terramycin (oxytetracycline) were given to all juvenile coho salmon at LPW during this period. The first treatment, from August 4 to 13 was given because of small numbers of dying fish and to attain high antibiotic blood levels in HR-0 fish when this population was divided on August 13. Oxytetracycline was mixed in Oregon Moist Pellets at a rate of 4 g per 100 pounds of fish per day for the 10 days. The second treatment, from October 9 to 18, using commercially-mixed 4% TM-50 in the food, was given to attain high antibody levels in the fish population during scheduled fish handling and tagging activities in late October.

Feeding and Food Conversions

Juvenile coho salmon were fed to satiation 8 to 14 times per day during the Phase 1 and Phase 3 periods and generally 2 to 6 times during the Phase 2 period. On bright sunny days in July, August, and early September and again in April and May, there was frequently a noticeable reduction in feeding behavior responses during the middle of the day. When these conditions occurred there was generally a dramatic increase in feeding behavior responses during the evening crepuscular period.

In previous years, efforts to determine conversion ratios of food fed to increases in fish weight frequently were not practical due to significant losses of fish during test periods. Food conversion ratios for juvenile coho salmon in the two horizontal raceways was determined for two time periods in 1975 when population numbers remained stable. During 17 days in August the food conversion ratio was 1.37:1 and during the 30 days of September it was 2.35:1 (Table 4).

PHASE 2 CULTURE

The overwinter period at LPW is characterized by low water temperatures, slow growth of fish and frequently severe icing conditions or heavy snowfalls that create hardships on maintaining fish culture units. Snowload and icing became serious concerns relative to the operation of floating raceways (Figure 12) especially in the two horizontal units (HR-1 and HR-2).

The vertical raceways (VR-1 and VR-2) functioned well under all weather conditions experienced. Due to their relatively small surface area, icing was never a problem even with low (30 gpm) flows. The 8 ‰ salinity water flowing through the vertical raceways had a higher temperature and lower freezing point than the fresh water flowing through the horizontal raceways. Average temperatures of water flowing through the vertical and horizontal raceways and from a 2-m depth in the estuary during the overwinter period are compared in Figure 11. The flotation collars from which vertical raceways were suspended provided sufficient buoyancy to maintain adequate freeboard even before accumulations of heavy snowfalls could be removed from surrounding walkways.

During prolonged intervals of subfreezing temperatures and heavy snowfall, ice began forming on the horizontal raceways due to their relatively large exposed surface area and colder fresh water. Waterflow to the horizontal raceways was increased to 400 gpm (R=0.9) and re-plumbed to maximize surface agitation in an attempt to minimize ice formation. A compressed air system was also installed inside the horizontal raceways to provide a bubble curtain for additional agitation. The spruce-log flotation structure from which the horizontal raceways were suspended did not provide adequate buoyancy, and snowloads on the walkways frequently reduced freeboard to critically low levels. This

Table 4. Estimated food conversion of 1974 brood coho salmon in horizontal raceways^{1/} at Little Port Walter during two time periods.

Time Period	Pounds of Fish		Pounds Gained	Pounds of food	Conversion Ratio Food/Fish
	Start	Finish			
1. <u>August 13 to 31</u> (17 days)	974	1688	714	994	1.39 : 1
2. <u>September 1 to 30</u> (30 days)	1688	2563	875	2053	2.35 : 1

^{1/}HR-1 and HR-2 combined.

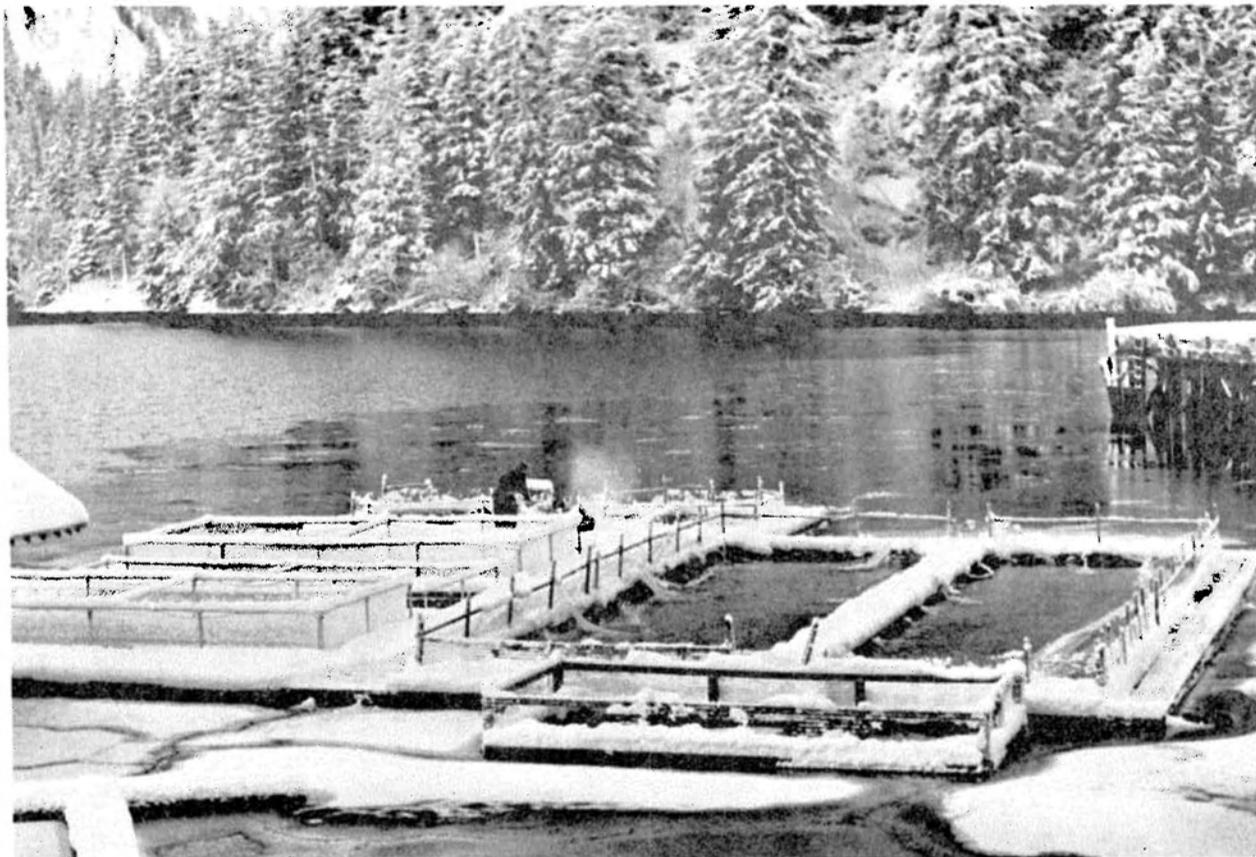


Figure 12.--Winter conditions at Little Port Walter experimental estuarine culture facility. Inflow water on floating horizontal aceways has been dispersed to maximize surface agitation. Snowload on flotation structures must be removed frequently.

situation required vigilance in snow removal from walkways (even at night with heavy snowfalls). At times a real threat existed that the surface of the horizontal raceways would freeze. Should this occur during a period of heavy snowfall, the additional area (frozen surface of the raceways) accumulating snow weight combined with the minimum buoyancy margins of the flotation structure, could have sunk the raceways to a point where the fish populations in them could have escaped. The seriousness of this possibility was modified only by the relatively mild 1974-75 winter period.

All six populations were sampled for growth, and biomass estimates of numbers were made at the end of the overwinter period (March 23 to April 5). During this time, lots of fish from some of the populations were marked with coded wire tags and released to evaluate the effects of the particular culture treatments on ocean survival from early spring releases of smolts. Some of the overwinter populations were subdivided into new populations for Phase 3 (spring) culture while other portions were released unmarked. In one case (NP-2) the overwinter population was maintained intact throughout the spring period (except for processing for growth and survival and transferring to a clean net pen).

Beginning on March 15 all populations were started on a 10-day ration of oxytetracycline-medicated food to elevate antibiotic blood levels during the forthcoming period of handling, moving, and marking fish. This is a prophylactic measure against possible disease complications arising from stress during handling of fish.

Two additional flowthrough formalin treatments were given to the populations in HR-1 and HR-2 because intense "flashing" behavior suggested probable external protozoan infections. The first of these treatments was given to both populations on November 28. The second was given to HR-1 on March 26 and to HR-2 on April 2. We will discuss later the implications of these second treatments as they relate to protozoan infestations and rapid adaptations of juvenile coho salmon to fall seawater.

Growth and Survival

Growth rates in the six populations averaged about 0.1% in the net pen, 0.2% in the horizontal and 0.3% in the vertical raceway groups during the overwinter period (Table 5). These rates only partially correlated with water temperatures. While the HR and VR populations (fresh water and 8 ‰ salinity) were positively correlated with increases in temperature, the slower growth in the NP populations (29 to 31 ‰ salinity) was associated with the warmest temperature (Figure 11). Initial and final mean weights of fish during this period plotted against cumulative temperature units for the three types of culture treatments (HR, VR, and NP) shows the slope of the rate of growth relative to temperature (Figure 13).

Table 5. Comparisons of lengths, weights, numbers of fish, growth, and survival rates in six cultured populations of 1974 brood coho salmon at LPW between October 20, 1975 and April 5, 1976.

Population	Sample date	Mean fork length (mm)	Mean weight		Population size		Rate of growth (percent)	Survival (percent)
			fish per pound	grams per fish	pounds	number (thousands)		
HR-1	Oct. 20	91.2	53.6	8.47	1161.0	62.2		
	Dec. 30		48.4	9.38			0.1	
	April 1	101.6	40.5	11.21	1494.3	60.5	0.2	97.3
HR-2	Oct. 21	90.7	53.9	8.42	1173.0	63.2		
	Dec. 30		48.4	9.38			0.2	
	April 5	101.5	39.2	11.58	1509.1	59.2	0.2	93.6
VR-1	Oct. 22	87.5	56.5	8.04	168.4	9.5 ^{1/}		
	April 1	106.0	37.1	12.24	256.1	9.5 ^{1/}	0.3	100.0
VR-2	Oct. 22	87.3	57.0	7.96	167.6	9.5 ^{1/}		
	April 1	104.3	34.6	13.12	268.0	9.3 ^{1/}	0.3	97.9
NP-1	Oct. 21	91.0	53.9	8.42	185.6	10.0 ^{1/}		
	Mar. 30	98.9	46.1	9.85	199.2	9.2 ^{1/}	0.1	92.0
NP-2	Oct. 21	91.0	53.3	8.52	187.8	10.0		
	Mar. 23	97.1	50.0	9.08	192.0	9.6	> 0.1	96.0

^{1/} Individual counts of fish terminating VR-1, VR-2, and NP-1 populations were 9511, 9271, and 9174.

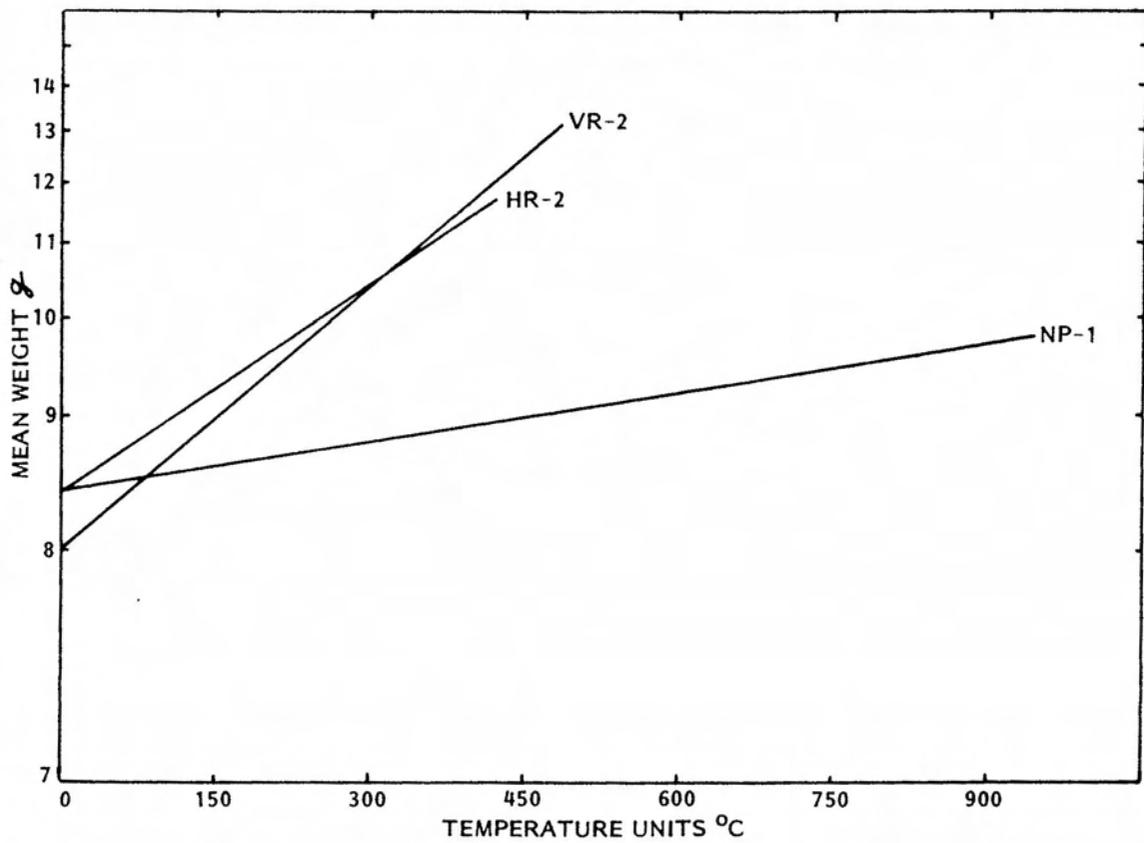


Figure 13.--Cumulative centigrade temperature units in three populations of juvenile coho salmon compared with mean weight of fish in each population at the beginning and end of the Phase 2 (overwinter) culture period.

We interpret these data to indicate that the overwinter temperature and potential growth advantage in full seawater was overridden by a low level osmoregulatory stress in the net pen populations. The 8.4- to 8.5-g fish that started these populations in October were generally in excellent health and well above the presumed 5- to 6-g threshold size required for successful overwinter seawater culture at LPW. The 92 and 96% survival of these fish (Table 5) is the best recorded to date for overwinter culture at LPW in high salinity net pens. The 3 to 5 fish per day overwinter mortality in these populations is thought to represent individuals succumbing to chronic osmoregulatory difficulty.

Although growth rates during Phase 2 culture were low compared to those in the Phase 1 period (Tables 3 and 5), they are nevertheless important, especially the 0.3% per day rate in the vertical raceways. This rate, over the 162-day interval from October 22 to April 1, allowed an overall 58% increase in mean weight of fish in the VR populations. This, during a period when normally little growth is thought possible, is a significant gain. More careful study of salinity-temperature interactions on growth of different sizes of juvenile coho salmon could lead to a precise culture strategy designed to maximize overwinter advantages of seawater over freshwater temperatures.

The overwinter loss of fingerlings from the horizontal raceways (about 5700 fish, Table 5) was due to vertebrate predators, principally birds. Some fish were lost due to entanglement in cover nets and to stranding on ice. Predation by water ouzels (*Cinclus mexicanus*), gulls (*Larus* spp.), and kingfisher (*Megaceryle alcyon*) was observed. Land otter (*Lutra canadensis*) were seen in the area but were never seen attempting to enter any of the culture units. Mink also were not thought to be a problem during this period.

Much of the overwinter loss in the horizontal raceways was believed due to ouzel predation because this bird could readily move back and forth through meshes of the net covers (which were designed to exclude gulls, kingfishers, mink, and otter yet not allow snow to accumulate during heavy snowfalls). Ouzels were territorial and one bird would aggressively chase away others of its own species. Based on carcasses of ouzel-killed fingerling coho salmon along the walkways, most of the fish caught were <70 mm.

Overall survival during the Phase 2 period, based on an estimated 164,400 fish at the beginning and 157,300 fish at the end of the period, was 95.7%.

Marking, Early Spring Releases, and Population Evaluation

Basic details of the marking procedure with coded wire tags previously described for LPW (Heard, Martin, and Wertheimer, 1975) were followed with 1974 brood coho salmon. Marking and other fish processing

activities were coordinated to minimize moving and handling of fish. For example, if part of one population was scheduled for marking with coded wire tags, another part for releasing unmarked, still another part for continued culture or subdividing into a new population treatment, these events were simultaneously organized to avoid repeated handling of the same fish.

One modification from previous marking and handling procedures involved the length of time fish were held in the prerelease holding net. In earlier years we usually held fish, just prior to release (after marking and final processing), for 48 h in a 12- by 12- by 12- by 1/4-inch-mesh prerelease net pen in the estuary. These earlier releases all involved fish from estuarine net-pen culture that had been exposed to full-strength seawater for extended periods. In these cases holding time was designed mostly to measure deaths of fish injured during final handling. Because 1974 brood coho salmon involved groups that were cultured in fresh water and intermediate salinity water up to the time of final marking or handling, and because some questions arose concerning rapid adaptations of these groups to full-strength seawater (including possible adverse effects from protozoan infestations), the prerelease holding time was extended in some cases to as long as 168 hours (7 days). This provided for better assessment of initial mortality and of behavioral changes associated with these important events. Actual releases of fish were made in darkness by allowing the holding net to sink.

A change in procedure of marking with coded wire tags involved the use of fresh water or intermediate saline water in anesthetic baths, in the quality control device (QCD) of the tagging unit, and in the anesthetic recovery baths for those fish originating from these same culture environments. Earlier we used a small submersible pump to provide seawater for these purposes. Continued difficulty in correctly operating the QCD (separating tagged from untagged fish) with low water pressure from this pump led to using the QCD with a pressurized freshwater system.

Correct operation of the QCD is important in evaluating the presence of tags in fish and in determining the initial tag loss (shedding rate) once tags have been applied. Evaluation of the initial shedding rate is essential in determining how many fish released from each group actually have tags when liberated. Tag-shedding is a function of the quality of the tagging operation. Related primarily to tag placement in the fish's snout, it is correlated with many variables including size of fish, size of headmold, needle penetration, and tag implantation depth. Tag-shedding rates were determined for five lots (from the total of 24) of 1974 brood juvenile coho salmon marked and released at or near LPW. Shedding rates ranged from 0 to 3.7% and averaged 1.9% over time intervals ranging from 0 to 54 days (Table 6).

Correction factors for prerelease shedding of tags were determined as follows. Shedding rates determined for the five specific lots in Table 6 were applied to those same groups except for the data 2/6 lot that indicated no shedding. Because some shedding is presumed to occur in all groups, the average rate of 1.2% used for all other tagged lots (not included in Table 6) was also used for the data 2/6 group. The 1.2% rate was derived from Table 6 with the data 2/15 group excluded. The data 2/15 group was excluded from computing this average because the 57-day prerelease holding period was not typical of other releases made at LPW and because no shedding rate was determined for this group shortly after tagging. The number of fish released in each tagged group, except as previously noted, was multiplied by the reciprocal of the average shedding rate, the tag retention rate ($=0.988$), to estimate the numbers of fish released with tags.

The following account is a summary of events associated with the population evaluations made at the end of the Phase 2 culture period. It covers a brief description of the disposition of each of the six overwinter populations including marked and unmarked releases and the development of new populations for Phase 3 culture.

A. VR-1 and VR-2

The comparison of survival of lake stock progeny (VR-1) to survival of EPR stock progeny (VR-2), to test for the possible occurrence of natural immunity against vibriosis, was terminated by counting the numbers of fish remaining in each population. The results, VR-1=9511 fish and VR-2=9271 fish, led to rejecting the hypothesis that VR-2 had any survival advantage over VR-1 under the culture conditions encountered. An important point here, of course, is that no vibriosis epizootic occurred. All fish in VR-1 were released unmarked while those in VR-2 were divided into four subgroups and treated as follows: one group of 2426 fish was marked with excised adipose and dorsal fins and released; one group of 2453 fish was marked ad+cwt (data 2/4) and released; one group of 2600 fish was designated as a new population, VR-3, for continued rearing at 8 ‰ salinity until the end of Phase 3 culture; and one group of 1596 fish was treated with formalin, held for two days, marked ad+cwt (data 2/8), and released.

A brief rationale for these marked releases includes: ad+dorsal group to provide a discreet, visible mark on some adults for possible continued breeding of the EPR stock (a dorsal mark only was not adequate because of some dorsal marks on juvenile coho salmon from nearby lake studies); ad+dorsal and ad+cwt groups released from the same population at the same time to provide information on comparative effects of these different marks on ocean survival; and the two ad+cwt groups (data 2/4 and 2/8) to provide information on possible beneficial effects of formalin treatments on ocean survival (the latter point is related to presumed protozoan infestations and their reported adverse effects on seawater acclimation).

Table 6. Numbers of fish examined and percent of those shedding coded wire tags from five groups of juvenile coho salmon at LPW in 1976.

Code number and population origin	Tagging date	Days from tagging to examination for shed tags	Numbers of fish examined	Number without tags	Percent shed tags
Data 2/4: VR-2	March 28	1-2	171 ^{1/}	2	1.2
Data 1/12: NP-1	March 28	0	137	4	2.9
Data 2/8: VR-2	April 3	0	244	4	1.6
Data 2/6: HR-4	May 2	2	280	0	0
Data 2/15: VR-4	June 29	57	325	12	3.7
Total			1157	22	1.9
Total excluding data 2/15			832	10	1.2

^{1/} Includes 19 fish that died in the prerelease holding period.

B. NP-1 and NP-2

All NP-1 fish were marked ad+cwt (data 1/12) and released. All NP-2 fish were transferred to a clean 12- by 25- by 12-foot by 1/4-inch-mesh net pen for continued culture. Both of these groups related to the fall release (data 1/11) of age 0 fish at the end of the Phase 1 culture period.

C. HR-1 and HR-2

The formalin treatment of HR-1 on March 26 preceded by 5 days any processing of fish in this population. Beginning on March 31, 9772 fish from HR-1 were marked ad+cwt (data 1/13) and released as an early freshwater culture group. A group of 3060 fish from HR-1 were transferred to a 12- by 12- by 12-foot by 1/4-inch-mesh net pen and designated as a new population, NP-3, to be cultured during the Phase 3 period. A total of 47,510 unmarked fish were released from HR-1 between March 31 and April 3.

On April 2, 2460 fish were removed from HR-2 (before the formalin treatment on the same date for the remainder of the population) and placed in a 12- by 12- by 12-foot by 1/4-inch-mesh net pen for a comparative seawater acclimation test with several other groups. A total of 29 fish from this test group died over the next 7 days; the remaining 2431 fish were released unmarked on April 9. HR-2 was terminated on April 5 when the remaining fish were divided (by weight) into two equal groups and designated as new horizontal raceway populations HR-3 and HR-4. Both groups were cultured during the Phase 3 period; HR-3 was continued as a freshwater population with a flow of about 225 gpm, HR-4 became a hypertonic intermediate salinity population with an average salinity of about 17 ‰ and a flow of about 200 gpm.

All marked and unmarked releases of 1974 brood coho salmon made from the estuarine culture facility at the end of the Phase 2 period are summarized in Table 7.

Adaptations to Seawater

As previously discussed, releases of cultured salmon at LPW are generally made from release net after a post-handling or post-marking holding period. All net pens now used at LPW are relatively deep (10 to 12 feet) and most of the volume is in 29 to 31 ‰ seawater (below the variable surface halocline). The initial transfer of any new group of fish into a net pen, either for release or continued culture, serves as a crude bioassay of the osmoregulatory capability of the fish. The deaths of fish in the first few days following transfer provide a cumulative measure of the effects of stress or injury during handling and possible stress of rapid acclimation to full seawater.

Table 7. Summary of marked and unmarked releases of 1974 brood juvenile coho salmon released at LPW at the end of the Phase 2 culture period (early spring 1976).

Date of Release	Number Released ^{1/}	Mean weight fish per pound grams per fish		Marks, treatments, and population origin
March 30	2426	34.2	13.3	Ad+dorsal; EPR stock discrete mark; VR-2
March 30	2453	34.4	13.2	Ad+cwt, data 2/4; Phase 2, 8°/oo salinity; VR-2
April 5	1596	34.7	13.1	Ad+cwt, data 2/8; formalin treated 8°/oo salinity; VR-2
March 31	9127	46.1	9.8	Ad+cwt, data 1/12; Phase 2, 29-31°/oo salinity; NP-1
April 3	9906	40.5	11.2	Ad+cwt, data 1/13; overwinter fresh water; HR-1
April 1	9511	37.1	12.2	Unmarked; terminate; VR-1
March 31- April 3	47510	40.5	11.2	Unmarked; terminate; HR-1
April 9	2431	39.2	11.6	Unmarked; seawater acclimation test; HR-2
Total marked releases:		25,493		
Total unmarked releases:		59,452		

^{1/} Numbers are adjusted for all known post tagging-post handling deaths; they do not include estimates for prerelease shedding of wire tags.

Variations in behavior of fish recently transferred into net pens are also indicative of cumulative effects of stress and osmoregulatory problems. Rapid return to pre-transfer feeding levels and use of the entire net pen volume are behavioral clues to successful acclimation. Fish in moderate osmoregulatory stress commonly remain tightly grouped in the bottom of the net pen and show little or no interest in food.

Most groups of coho salmon released at LPW in the past have shown 24- and 48-h prerelease mortalities (after marking or handling) of >0.2%. When pre-release mortalities increased dramatically in late March 1976 (to as high as 3.8% in 72 h in one case), we suspected infections of a protozoan ectoparasite, possibly Costia. Our suspicions were based on intense flashing behavior in some populations and on reported difficulties in adapting to seawater caused by costiasis and other protozoan infections (Wood, 1974). Although flashing behavior was not noted in VR-2, the initial population with high prerelease losses, flashing was abundantly evident in both horizontal raceways which provide better opportunity for viewing this behavior than the vertical raceways.

Samples of horizontal (fresh water) and vertical (8 ‰ saline) raceway-cultured fish were sent to the Pathology Laboratory on March 24 for analysis for bacterial and parasitic infections. Gill tissue was specifically examined for Costia. No bacterial or protozoan isolates were found. However, the extent of flashing in HR-1 was greatly reduced by formalin treatment on March 26. A second sample taken from HR-2 on April 2 and examined for ectoparasites proved to have a moderately heavy infection of the protozoan Trichodina. No Trichodina were observed from gill tissues but they were common on scrapings from the body wall. This sample was taken just before a formalin treatment in HR-2 that also greatly reduced flashing behavior.

During the culture history of 1974 brood coho salmon at LPW, we presume the periodic build-up of flashing behavior in the horizontal raceways could have been caused by Trichodina. Periodic formalin treatments also appeared to be effective in reducing these infections. Whether Trichodina, by impairing rapid transfer to seawater, was causing the increased mortality of juvenile coho salmon in prerelease holding nets was another question.

Two of three groups of fish from VR-2 population were marked and released without formalin treatments (ad+dorsal and data 2/4) while the third (data 2/8) was formalin-treated before marking and release (Table 7). The 48-h prerelease mortality of these groups ranged from 2.2 to 2.4% with no measurable advantage over the formalin treatment.

A second test was conducted on prerelease mortality of fish treated and not treated with formalin. In this case both groups were transferred from fresh water after en masse weighings to estimate numbers; individual fish were not handled or marked. The formalin-treated group (NP-3) originated from HR-1. The non-formalin group (control) was taken from HR-2

just before it was formalin-treated. Both showed dramatically lower mortality rates in seawater than the VR-2 groups. Over a 7-day period the formalin-treated group had one-half the mortality rate of non-treated group (0.6 versus 1.2%) indicating, in this case, a beneficial effect from the formalin treatment on fish exposed to seawater. A binomial test of significance indicate the formalin-treated fish survived better (at about the 95% level of confidence) than the non-formalin-treated fish after 48, 72, and 168 h. Details of these two tests along with prerelease mortality rates of most of the marked lots of 1974 brood coho salmon released at LPW are reviewed in Table 8.

While important differences limit the extent to which the two above tests can be compared (neither Trichodina nor flashing was seen in VR fish; without being anesthetized or marked, the HR fish in the second test were subject to less handling stress), some parallels were noted. Behavioral responses were similar in both tests when fish were transferred to seawater net pens. Fish schooled tightly in the bottom of the nets and did not feed. Formalin-treated HR-1 fish (which became NP-3), although having the lowest mortality rate of any late March-early April groups transferred to seawater (Table 8), did not resume normal feeding behavior for about 10 days. Other fish processed in a similar manner but retained in fresh water (HR-3) or that underwent a less dramatic salinity change (HR-4=18 ‰) were feeding normally within 1 to 2 days after moving. In a related manner, Wedemeyer (1972) found that under mild handling stress (roughly comparable to our en masse weighings without handling individual fish), a minimum 24-h interval was required for blood chemistry of juvenile coho salmon handled in soft water to return to pre-stress conditions.

A review of all these factors led us to conclude that while in some cases Trichodina infections may have added to difficulty in rapidly adapting to seawater, there was an underlying lack of complete physiological readiness for abrupt transfer to seawater in late March-early April. Comparisons of ocean survival rates of adult returns from releases made during this time with releases made at other times will shed light on the significance of this conclusion.

Early Marine Observations

Following releases of juvenile coho salmon into the LPW estuary in late March-early April, 1976, many schools of several thousand fish were observed in the immediate vicinity for about 2 weeks. For a few days after the first releases, the schools observed near the net pens showed behavior similar to that observed in the prerelease nets: they were tightly grouped with little or no feeding response.

During early April, kingfishers; red-breasted mergansers, Mergus serrator; common mergansers, Mergus merganser; pelagic cormorants, Phalacrocorax pelagicus; and harbor seals, Phoca vitulina, were observed

Table 8. Cumulative mortalities of 1974 brood coho salmon after 48, 72, and 168 h in seawater net pens at Little Port Walter following various procedural treatments.

Population origin and salinity Population ‰	Date into seawater	Treatments ^{1/}			Initial numbers into seawater	Cumulative Mortality numbers in () and percent			
		ad+cwt	other	F/NF		48 h	72 h	168 h	
HR-1 and 2	0	10/21/75	1/1T		NF	10021	(1) <0.1	(2) <0.1	--
NP-1	30	3/28/76	1/12		NF	9174	(74) 0.5		
VR-2	8	3/27/76		ad+D	NF	2523	(60) 2.4	(97) 3.8	--
VR-2	8	3/28/76	2/4		NF	2508	(55) 2.2	-	--
VR-2	8	4/03/76	2/8		F	1637	(40) 2.4	-	--
HR-1	0	3/31/76	1/13		F	9943	(37) 0.4	-	--
HR-1	0	4/02/76		UM	F	3060	(9) 0.3	(15) 0.5	(18) 0.6
HR-2	0	4/02/76		UM	NF	2460	(16) 0.7	(25) 1.0	(29) 1.2
HR-3	0	5/02/76	2/5		NF	2518	(4) 0.2	-	--
HR-4	17	5/02/76	2/6		NF	2506	(1) <0.1	-	--
HR-3	0	5/19/76	2/9		F	2565	(3) 0.1	(6) 0.2	(6) 0.2
NP-2	30	5/26/76	1/14		NF	9136	(13) 0.1	-	--
NP-3	30	5/27/76	2/7		NF	3041	(1) <0.1	-	--
VR-3	8	5/27/76	2/12		NF	2506	(18) 0.7	-	--

^{1/}All groups were weighed en mass prior to transfer into seawater; in addition groups with marks were anesthetized and fish individually handled; ad+cwt means excised adipose fin plus coded wire tag; 1/11, 1/12, etc. identifies tag codes; ad+D means excised adipose and dorsal fins; UM means unmarked; F indicates that a formalin treatment was given to the population within a 6-day period prior to transfer; NF means no formalin treatment.

Table 9. Comparisons of lengths, weights, numbers of fish, growths, and survival rates in five cultured populations of 1974 brood coho salmon at Little Port Walter between March 23 and May 29, 1976.

Population	Sample date	Mean fork length (mm)	Mean weight		Population size		Rate of growth (percent)	Survival (percent)
			fish per pound	grams per fish	pounds	numbers (thousands)		
HR-3	April 5	101.5	39.1	11.61	721.3	28.2 ^{1/}		
	May 2	107.1	35.6	12.75		-2.5 ^{2/}	0.4	
	May 19	109.1	34.2	13.27		-2.6 ^{2/}	0.2	
	May 29	111.3	32.5	13.97	680.8	22.1	0.5	96.5
HR-4	April 5	101.5	39.1	11.61	727.0	28.4 ^{3/}		
	May 2	105.7	34.5	13.16		-2.5 ^{3/}	0.5	
	May 28	118.8	28.7	15.82	855.3	24.5	0.7	95.1
VR-3	April 1	104.3	34.9	13.01	74.4	2.6 ^{4/}		
	May 27	119.1	27.2	16.69	92.3	2.5 ^{4/}	0.5	96.2
NP-2	Mar. 23	97.1	50.0	9.08	192.0	9.6 ^{4/}		
	May 25	111.4	34.4	13.20	265.6	9.1 ^{4/}	0.6	94.5
NP-3	April 1	101.6	40.5	11.21	75.5	3.1 ^{4/}		
	May 27	118.0	27.2	16.69	112.2	3.0 ^{4/}	0.7	96.8

^{1/} 2518 removed for mid-spring release, data 2/5.

^{2/} 2565 removed for Toledo Harbor 11-day imprinting, data 2/9.

^{3/} 2506 removed for mid-spring release, data 2/6.

^{4/} Individual counts of fish terminating VR-3, NP-2, and NP-3 populations were 2506, 9136, and 3041.

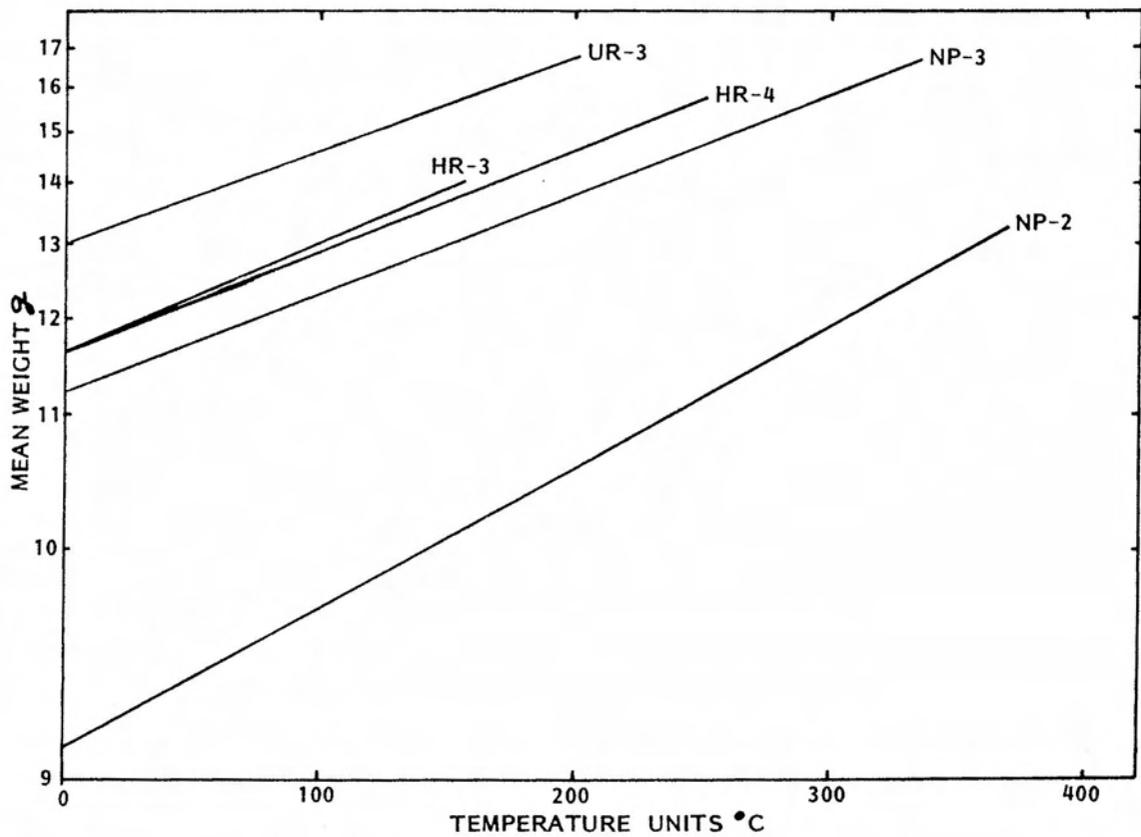


Figure 14.--Cumulative centigrade temperature units in five populations of juvenile coho salmon compared with mean weight of fish in each population at the beginning and end of the Phase 3 (spring) culture period.

feeding on fish in LPW Bay that included the recently released coho salmon.

On April 3, when about 25 mergansers and 3 cormorants were observed feeding in the area, a red-breasted merganser and pelagic cormorant were shot in the inner portion of LPW Bay to examine their stomach contents. The merganser stomach contained six digested fish at least one of which was a juvenile coho salmon. The cormorant stomach contained three freshly-injected coho salmon, each about 100 mm long.

On April 14, juvenile coho salmon were observed feeding at the surface of the inner and outer portions of LPW Bay on small decapod crustaceans (possibly mycids). On the same date, divers observed large schools of coho salmon smolts near the husbandry floats just off the bottom in 35 to 45 feet of water. These fish appeared to be foraging on epibenthic organisms. On April 15, coho salmon smolts were seen feeding on pink salmon fry and mycids near the surface of LPW estuary. By April 17, divers could not find large numbers of smolts around or beneath the raceway and net pen floatation structures, although smaller schools (>100 fish per school) were still present.

PHASE 3 CULTURE

Five populations of 1974 brood coho salmon were cultured during the Phase 3 period: NP-2 and NP-3 (29 to 31 ‰ salinity); VR-3 (8 ‰ salinity); HR-3 (fresh water); and HR-4 (17 ‰ salinity). This period is characterized by increasing photoperiod, temperature, and growth.

Some variations developed with intermediate saline flow to HR-4. This was due to positioning of the venturi device relative to the main manifold on the bay floor and the flexhose between the manifold and raceway. The salinity of water flowing through HR-4 occurred in two ranges, initially at 19 to 21 ‰ and subsequently at 14 to 16 ‰; it averaged about 17.5 ‰ for the 2 months this population was maintained.

Growth and Survival

Growth rates ranged from 0.3% (HR-3) to 0.6% (NP-3) per day during the Phase 3 culture period (Table 9). Growth was essentially temperature-dependent regardless of salinity. Mean weight of juvenile coho salmon at the beginning and end of this period plotted against cumulative temperature units in each of the five populations shows (Figure 14) that growth was parallel to temperature.

Survival during the Phase 3 period, while generally high in all units (Table 9), was complicated by the fact that mink had access to fish in some units but not others. It was discovered in May that mink could pass through the small-mesh net covers over HR-3, HR-4, and VR-3.

The cumulative loss of 2,500 fish in these three units is believed due to mink predation. Very few dead or distressed fish were observed in these units, and, other than an increase in flashing behavior during mid-May in HR-2 and HR-3, those populations were thought to be in excellent health. Four mink, all males, were livetrapped on the floats around these three units and removed from the LPW area in late May. A similar influx of mink activity occurred in the same interval during 1975, suggesting a seasonal pattern to mink predation problems at LPW.

There was no indication that mink could gain access to fish in NP-2 or NP-3 because of the elevated top of the webbing (in effect creating a fence) and the arrangement of the aluminum frames (to which net pens were attached) in the flotation collars. We observed a low level loss of fish in NP-2 during Phase 3 culture apparently from continued chronic osmoregulatory stress. This contrasted with NP-3 (established in early April) where few dead fish and no osmoregulatory stress was seen.

Overall survival during the Phase 3 culture period, based on an estimated initial 71,900 fish and 68,800 fish at end of the period, was 95.7%. These data take into account groups of fish removed from HR-3 and HR-4 in early and mid-May.

Population Evaluations and Phase 3 Releases

A mid-spring release of smolts marked with coded wire tags was made from fresh water (HR-3, data 2/5) and 17 ‰ saline culture (HR-4, data 2/6) on May 4 at LPW. These releases were designed to compare ocean survivals with similar releases made earlier (from fresh water) and later (from fresh water and 17 ‰ salinity).

Both HR-1 and HR-2 populations were treated with formalin on May 17 for flashing that had again increased to a moderately intense level. The presumed causative protozoan, Trichodina, apparently can survive equally well in fresh water and 17 ‰ saline water. Recently a marine form of Trichodina has been reported from coho salmon in saltwater pens in Washington (Al Didier, personal communication). Flashing behavior in HR-3 and HR-4 almost disappeared following the formalin treatments.

On May 18, we began a 10-day feeding schedule for all fish with food medicated with oxytetracycline. The purpose was to raise resistance to possible disease complications from stress associated with terminal handling and processing at the end of May.

The following is an account of the terminal population evaluations and releases made from the five groups of 1974 brood coho salmon cultured at LPW during the Phase 3 period. All marked and unmarked releases at LPW during this period are summarized in Table 10.

Table 10. Summary of marked and unmarked releases of 1974 brood juvenile coho salmon released at Little Port Walter at the end of the Phase 3 culture period, spring 1976.

Date of Release	Number Released ^{1/}	Mean weight		Marks, treatments, and population origin
		fish per pound	grams per fish	
May 4	2514	35.6	12.8	Ad+cwt; data 2/5: mid-spring fresh water; HR-3
May 4	2505	34.5	13.2	Ad+cwt; data 2/6: mid-spring, 17°/oo salinity; HR-4
May 28	9123	34.4	13.2	Ad+cwt; data 1/14: Phase 2-3; 29-31°/oo salinity; NP-2
May 28	2509	28.7	15.8	Ad+cwt; data 2/13: Phase 3; 17°/oo salinity; HR-4
May 29	2488	27.2	16.7	Ad+cwt; data 2/12: Phase 3; 8°/oo salinity; VR-3
May 29	3040	27.2	16.7	Ad+cwt; data 2/7: Phase 3; 29-31°/oo salinity; NP-3
May 30	2559	32.5	14.0	Ad+cwt; data 2/9: Imprint Toledo Harbor 11 day; HR-3
44 May 30	2551	32.5	14.0	Ad+cwt; data 2/11: Imprint Toledo Harbor 1 day; HR-3
May 30	2574	32.5	14.0	Ad+cwt; data 2/10: Imprint Control, Phase 3; fresh water; HR-3
May 28	21348	28.7	15.8	Unmarked; Terminate, 17°/oo salinity; HR-4
May 29	14362	32.5	14.0	Unmarked; Terminate, fresh water; HR-3
Aug. 25 ^{2/}	2584	9.4	48.3	Ad+cwt; data 2/15: Delayed, summer release, fresh water; VR-4
Total marked releases		32,447		
Total unmarked releases		35,710		

^{1/}Numbers are adjusted for all known post tagging-post handling deaths; they do not include corrections for prerelease shedding of wire tags.

^{2/}This group of fish was not released in the Phase 3 culture period; it is discussed under delayed release in the text.

A. HR-3: Imprinting-Homing Transport and Control Groups

Two groups of smolts from freshwater culture in HR-3 were used in an imprinting-homing study which involved transporting fish to 12- by 12- foot by 1/4-inch-mesh net pens at Toledo Harbor, a small bay one mile south of LPW (Figure 1). The purpose of this previously-described study (Heard, Martin, and Wertheimer, 1975) was to evaluate the possible imprinting effects of exposing coho salmon smolts to two intervals of time at Toledo Harbor. Releases of age 1 1974 brood smolts were designed to compliment knowledge gained from earlier releases at Toledo Harbor of age 1 1972 smolts.

Principal differences in releases made from 1972 and 1974 brood coho salmon at Toledo Harbor included the numbers in released lots and the culture history prior to release. The numbers of 1974 brood releases were just over 2500 fish per lot versus 1972 brood releases of about 10,000 fish per lot. The 1974 brood releases were cultured in fresh water prior to saltwater exposure at Toledo Harbor while the 1972 brood releases were cultured for 7 months in saltwater net pens at LPW before transport to Toledo Harbor. In both broods, lots of marked coho salmon were held 11 days and 1 day at Toledo Harbor at the same time of year before release. Control lots were also released at the same time with both broods at LPW.

An important aspect of both of these groups of releases at Toledo Harbor is the fishless stream that flows into the bay. The stream is about one-fourth to one-third the size of Sashin Creek (both in drainage area and discharge) and has no fish because of an impassable falls just at the head of tidewater. The net pens from which coho salmon releases were made at Toledo Harbor were anchored in close proximity to the stream in both years.

The principal purpose of the two releases made in 1976 was to investigate the possible significance of the initial marine experience (saltwater exposure) on the imprinting process. To produce meaningful data and to relate to the available earlier information, the releases were made at the end of May, a time when imprinting is known to occur. On May 19, a lot of 2565 fish from HR-3 was marked ad+cwt (data 2/9) and transported in covered tubs to the net pen in Toledo Harbor. On May 29, a second lot of 2555 tagged fish (data 2/11) was also transported in covered tubs to the Toledo Harbor net pen. The 11-day and 1-day exposure lots were both released at 2200 hours on May 30. A control lot of 2574 fish (data 2/10) also tagged from HR-3 was released at the same time at LPW.

A group of 2600 fish was removed from HR-3 on May 29 and transferred into a new vertical raceway and designated population VR-4. This new population was scheduled for continued freshwater culture and a delayed release in late summer. It was related to other releases to be made with 1975 brood coho salmon cultured at LPW from accelerated and

ambient temperature regimes. Specific details of 1975 brood coho salmon culture at LPW will be covered in another report. Until VR-4 was established for freshwater culture, all earlier culture in vertical raceways at LPW with 1974 brood coho salmon had been at 8 ‰ salinity (VR-1, VR-2, and VR-3).

The remaining 14,362 coho salmon in the parent HR-3 population were released unmarked at LPW on May 29.

B. HR-4

The HR-4 population was evaluated on May 28. One lot of 2510 fish was marked ad+cwt (data 2/13) for release at LPW to determine possible influence of 17 ‰ saline culture during the Phase 3 period on ocean survival. The remaining 21,348 fish from this population were released unmarked.

C. VR-3

The population in VR-3 was removed on May 27, marked ad+cwt (data 2/12), and transferred to a prerelease holding net. Adult returns from the 2488 fish released on May 29 will provide a comparison of ocean survival of coho salmon smolts raised since early fry stage (10 months) at 8 ‰ salinity with other groups.

D. NP-2

The terminal evaluation and marking of coho salmon smolts in NP-2 extended over a 3-day interval, May 24-26. Ocean survival of the 9123 fish released from this population (data 1/14) will be compared with the other two 10,000 fish lots originating in October, 1975 (fall release data 1/11 and NP-1, data 1/12 released on March 31) as well as other groups of marked releases. Culture history of the NP2 population parallels the "standard culture treatment" of many releases from earlier broods of coho salmon smolts raised at LPW.

E. NP-3

The population NP-3 was cultured during the Phase 1 and Phase 2 periods in fresh water, then during the 2-month Phase 3 period in a seawater net pen at 30 ‰ salinity. Return rate of the 3040 smolts marked (data 2/7) and released will be compared with adult return rates of other groups.

Adaptations to Seawater

Fish groups transferred to prerelease seawater net pens after processing and marking during Phase 3 culture had lower mortality rates than those handled in a similar way at the end of Phase 2 culture (Table 8). The two mid-spring groups from HR-3 and HR-4 had 48-h prerelease mortality rates of 0.2% (data 2/5) and > 0.1% (data 2/6), respectively. The HR-4 group transferred from 17 ‰ saline water showed no stress behavior and began feeding actively within 3 or 4 h after recovering from the anesthetic. The HR-3 group from fresh water showed minor initial signs of osmoregulatory stress by not feeding and by remaining tightly grouped in the bottom of the net pen. Before the end of the 48-h, post-tagging, prerelease, holding period, however, the HR-3 fish were feeding and behaving normally.

The data 2/9 freshwater group marked from HR-3 on May 19 and introduced to seawater at Toledo Harbor showed no signs of stress and was actively feeding on the day of tagging and transport. The 168-h (7-day) mortality rate in this group of fish was 0.2% (Table 8).

Fish cultured in 8 ‰ saline water again showed the most severe reaction to transfer into seawater. The VR-3 population (data 2/12) had a 48-h mortality rate of 0.7% (Table 8). A binomial test for significance at the 95% level indicated this mortality rate was higher than all other groups during the Phase 3 culture period. This 48-h 0.7% rate, however, was also significantly lower than the 48-h mortality rates experienced by 8 ‰ saline cultured fish at the end of the Phase 2 period (VR-2 data 2/4, 2/8, and ad+dorsal groups).

No postmarking behavioral observations could be made on VR-3 fish because they were transferred to a prerelease holding net with another group of fish (NP-3, data 2/7). Dead fish from the two groups were separated by coded wire tag identification.

Delayed Release

The freshwater population VR-4, established at the end of Phase 3 culture, was maintained throughout June, July, and most of August. This period includes much of the primary growth interval associated with the Phase 1 culture period. However, we have conceptionally related Phase 1 culture to age 0 and not older age fish. While age 1 coho salmon cultured through a second Phase 1 period would have favorable growth conditions, the final end product of this extended culture, the adult return rates, specific location of adult returns, or some other benefit (perhaps unique benefit) should justify the additional cost of age 1 delayed-release smolts over the cost of age 1 smolt releases made at an earlier date.

The concept of delaying releases of cultured smolts to achieve particular benefits has received considerable attention in the Puget Sound area of Washington in recent years (Moring, 1976). Among the benefits identified with these releases are overall increases in marine survival and modified ocean migration patterns that make the fish more available to specific user groups. In Puget Sound, release of both coho and chinook salmon smolts made in July and August versus "normal" release in April or May tend to remain in local marine waters and contribute heavily to Puget Sound fisheries. A disadvantage identified with delayed releases of smolts is that returning adults tend to be smaller in size than those returning from smolts released earlier.

It remains, however, to be adequately documented just how well factors associated with delayed releases in Puget Sound apply to Alaska conditions. Preliminary indications are that some of the disadvantages of delayed smolt releases are occurring in Alaska (smaller size of returning adults) without concomitant advantages (increases in return rates of adults). At LPW mean weight of the delayed-release fish in population VR-4 increased from 14.0 g in late May to 18.8 g in late June and to 48.3 g in late August. Growth rates were 1.0% through late June and 1.7% during July and August.

This population was marked ad+cwt (data 2/15) on June 29 and returned to freshwater VR-4 for continued culture until the release date, August 25. Marking was completed ahead of the release schedule to avoid possible handling difficulties associated with (1) higher late summer temperatures, and (2) probable increases in sensitivity to handling, especially deciduousness of scales. A total of 2606 fish were tagged on June 29. The cumulative posthandling loss was four fish. Eighteen dead fish were removed from VR-4 in July and August; of these, 14 were killed by gilling themselves in the net cover over the top of the vertical raceway.

On August 25, 2584 fish were released; however, because this group of fish had an unusually high tag-shedding rate (Table 6), the estimated number of fish released with wire tags still in place was 2488. Samples of these fish were not checked for tag loss until the day of release and we do not know if the relatively high shedding rate is due to a group of fish poorly tagged initially or to the longer than normal time between tagging and checking for shedding.

SUMMARY OF MARKED AND UNMARKED RELEASES

The releases of marked and unmarked 1974 brood juvenile coho salmon at LPW from freshwater and estuarine culture in floating raceways and net pens is summarized (Table 11) for the three temporal phases of fry-to-smolt stage culture. These releases total 162,721 fish.

Table 11. Summary of 1974 brood coho salmon marked and unmarked releases from freshwater and estuarine culture in floating raceways and net pens at LPW during three culture periods.

Culture Period	Marked Fish Released		Unmarked Fish Released number
	number	specific lots	
Phase 1	10,019	1	-0-
Phase 2	25,493	5 ^{1/}	59,452
Phase 3	32,447	10 ^{2/}	35,710
Totals	67,559 ^{3/}	16	95,162

^{1/} Includes 2426 fish released with ad+dorsal mark (no coded wire tags).

^{2/} Includes the delayed release data 2/15 lot released on August 25, 1976.

^{3/} Of the total number of marked fish released, 65,133 originated from 15 coded wire tag lots; an estimated 782 of these have lost their tags and have ad-only marks based on prerelease shedding rates (1.2%, see text).

In addition to the 1974 brood releases made during 1975 and 1976, a total of 12,432 1975 brood age 0 coho salmon were also released at LPW in 1976. These 1975 brood releases included five separate marked lots cultured at ambient and various accelerated temperature regimes. All were cultured in fresh water before release. They are mentioned here only because some of these fish will likely impact the 1977 adult returns to LPW. Specific details of 1975 brood culture will be covered in a subsequent report.

A third group of juvenile coho salmon that will impact the adult returns to the LPW area in 1977 are the smolts that volitionally emigrated out of Osprey Lake following the planting of 276,000 fry in July 1975. The emigrants from Osprey Lake include 44,400 age 0 fish (11.0-g average) that left the lake in September and October in 1975 and 144,500 age 1 fish (9.0- to 12.0-g average) that left the lake in May and June 1976. Because of the close proximity of Osprey Lake and LPW (Figure 1) considerable intermixing of returning adults from these two areas may occur. Osprey Lake emigrants marked ad+cwt include four lots that total 8225 fish. These include one age 0 lot (data 2/3, 2403 fish) and three age 1 lots (data 1/15, 1472 fish; data 2/1, 2207 fish; and data 2/2, 2143 fish). Details of the Osprey Lake stocking will be covered in other reports.

Table 12 provides an overall summary of all of the coded wire tag lots of juvenile salmon released at or near LPW through 1976. Included are juvenile coho salmon releases made from 1972, 1974, and 1975 broods; brief comments on the purpose and treatment of each of the releases are provided. The numbers listed in Table 12 are corrected for estimates of prerelease shedding of tags; hence, for specific code lots of 1974 and 1975 brood coho salmon listed elsewhere in this report, the numbers listed in Table 12 are smaller generally by 1.2% (see earlier text and Table 6). The cumulative total number released from the 24 separate coded wire tagged lots of coho salmon from 1974 and 1975 broods at LPW and Osprey Lake is 85,691 fish. Based on our estimated tag-shedding rates, 1028 of these lost their tags and have ad-marks only.

FRY-TO-SMOLT SURVIVAL

The ultimate measure of success for any ocean ranching (release-recapture) program should rest on adult returns, including contributions to various fisheries, spawning escapements, or other spawner returns. It is, however, also important to measure the effectiveness of the programs, that provide juvenile salmon for ocean nursery areas.

Fry-to-smolt survival for 1974 brood coho salmon culture at LPW can be examined in several ways. Using the estimated 484,500 initial fry as a beginning base (Table 1), and by adding 188,900 emigrants from Osprey Lake to the 162,721 cultured releases (Table 11), fry-to-smolt stage survival is 72.6%. This does not consider an estimated 20,000 age 1

holdover fingerlings in Osprey Lake that will likely emigrate as age II smolts in 1977. Fry-to-smolt survival for Osprey Lake alone (planted fry through age 1 emigrant) is 68.7%. Using the initial 177,200 fry available for intensive culture in floating raceways and net pens as a base (beginning in mid-July 1975, Table 3), survival of fry to release was 91.8%.

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Table 12--Summary of NMFS (Agency 3) Binary Coded Wire Tags Released in Juvenile Salmon in Alaska through October 1976.

Code	Species/ Brood Year	Release Date	Release ^{1/} Location	Number ^{2/} Released	Mean Weight (gms)	Lot Treatments ^{3/}
1/2	Coho/72	April 9, 1974	LPW	2,276	4.4	Early; Age 1; NP
1/2	CH-1/71	May 20, 1973	LPW	7,261	14.2	Normal; Age 1; NP
1/3	Coho/72	April 8, 1974	LPW	9,351	7.6	Early; Age 1; NP
1/4	Coho/72	May 7, 1974	LPW	10,669	9.1	Middle; Age 1; FW; O/W NP
1/5	Coho/72	May 31, 1974	TH	10,806	12.0	Normal; Age 1; IH-11 day; NP
1/6	Coho/72	May 31, 1974	TH	10,485	12.0	Normal; Age 1; IH-1 day; NP
1/7	Coho/72	May 31, 1974	LPW	9,535	12.0	Normal; Age 1; IH-control; NP
1/8	Coho/72	June 30, 1974	LPW	9,989	21.0	Late; Age 1; NP
1/9	Coho/72	Sept. 7, 1974	LPW	1,946	65.0	Fall; Age 1; delay; NP
1/9	Sock/73	June 7, 1975	LPW	7,705	22.0	Normal; Age 1; O/W and Spring; NP
1/10	Pink/73	Sept. 6, 1974	LPW	9,905	19.7	Fall; Age 0; delay; NP
1/11	Coho/74	Oct. 24, 1975	LPW	9,899	8.4	Fall; Age 0; FW; HR
1/12	Coho/74	March 31, 1976	LPW	8,862	9.8	Early; Age 1; O/W; NP
1/13	Coho/74	April 3, 1976	LPW	9,772	11.2	Early; Age 1; O/W; FW/HR
1/14	Coho/74	May 28, 1976	LPW	9,014	13.2	Normal; Age 1; O/W & Spring; NP
1/15	Coho/74	May 1976	Osprey Cr.	1,454	~ 9.0	Early; Age 1; emigrant smolt
2/1	Coho/74	June 2, 1976	Osprey Cr.	2,181	~ 10.0	Normal; Age 1; falls bypass
2/2	Coho/74	June 3, 1976	Osprey Cr.	2,118	~ 10.0	Normal; Age 1; emigrant smolt
2/3	Coho/74	September 1975	Osprey Cr.	2,374	11.1	Fall; Age 0; emigrant "smolt"
2/4	Coho/74	March 30, 1976	LPW	2,424	13.2	Early; Age 1; 8°/oo; VR
2/5	Coho/74	May 4, 1976	LPW	2,484	12.7	Middle; Age 1; FW; HR
2/6	Coho/74	May 4, 1976	LPW	2,475	13.2	Middle; Age 1; O/W-FW; 17°/oo; HR
2/7	Coho/74	May 29, 1976	LPW	3,004	16.6	Normal; Age 1; O/W-FW; Spring; NP
2/8	Coho/74	April 5, 1976	LPW	1,577	13.1	Early; Age 1; 8°/oo; VR; formalin
2/9	Coho/74	May 30, 1976	TH	2,528	14.0	Normal; Age 1; IH-11 day; FW/HR
2/10	Coho/74	May 30, 1976	LPW	2,543	14.0	Normal; Age 1; IH-control; FW/HR
2/11	Coho/74	May 30, 1976	TH	2,520	14.0	Normal; Age 1; IH-1 day; FW/HR
2/12	Coho/74	May 29, 1976	LPW	2,458	16.6	Normal; Age 1; 8°/oo; VR
2/13	Coho/74	May 28, 1976	LPW	2,479	15.8	Normal; Age 1; O/W-FW; 17°/oo-HR
2/14	Coho/75	June 8, 1976	LPW	2,323	4.8	Normal; Age 0; accel.FW; 2 wk-8°/oo; VR
2/15	Coho/74	August 25, 1976	LPW	2,488	48.3	Late; Age 1; delay; FW/HR; VR

Table 12 --Continued

Code Data 1/ Data 2	Species/ Brood Year	Release Date	Release ^{1/} Location	Number ^{2/} Released	Mean Weight (gms)	Lot Treatments ^{3/}
3/1	Coho/75	August 25, 1976	LPW	2,565	3.6	Late; Age 0; ambient; FW; VR
3/2	Coho/75	August 26, 1976	LPW	2,414	16.9	Late; Age 0; accelerate; FW; VR
3/3	Coho/75	Oct. 25, 1976	LPW	2,486	15.9	Fall; Age 0; Int.accelerate; FW; VR
3/4	Coho/75	Oct. 26, 1976	LPW	2,496	7.8	Fall; Age 0; ambient; FW; VR

1/ LPW-Little Port Walter; TH-Toledo Harbor.

2/ Includes corrections for post tagging-prerelease mortalities and estimates of pre-release shedding of tags.

3/ Includes brief information on timing, age, culture history or purpose of release. Abbreviations include:

NP-net pen; FW-freshwater, O/W-overwinter; IH-imprinting homing; HR-horizontal raceway,
and VR-vertical raceway. Arbitrary assignments of timing categories include:

Early; March 15 to April 14
Middle; April 15 to May 14
Normal; May 15 to June 14
Late; June 15 to August 31
Fall; Sept. 1 to Oct. 31

