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**PEN REARING AND IMPRINTING
OF FALL CHINOOK SALMON**

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Pen Rearing and Imprinting of Fall Chinook Salmon

FINAL REPORT

BY

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ABSTRACT

Upriver bright fall chinook salmon (Oncorhynchus tshawytscha) are being reared in a backwater and a pond along John Day Reservoir to evaluate the benefits of rearing fish and releasing them off-station compared to traditional hatchery procedures. Fish reared in net pens at a density/feeding combination judged to be the economic optimum of those used during 1984 rearing trials exhibited good growth and smolt development. Size of fish averaged 112 fish/lb (4.0g/fish), ATPase activities ranged from 16.4 to 29.5 micromoles Pi/mg prot/hr at release and total mortality of fish was low among pens, ranging from 0.3 to 1.1%.

Poor growth and smolt development was observed in fish reared in a large barrier net, especially during the initial two weeks after stocking. In addition, mortality of fish in the barrier net was high (49%) in relation to any of the other treatments tested thus far. The combined effects of generally poor condition of fish at stocking, low zooplankton densities during the initial two weeks of rearing, and losses to predation were thought to be the primary causes of the slow growth rates and high mortality.

Unfed fish in pens utilized the available natural food base, but zooplankton densities were apparently not sufficient for growth, and may have been marginal for sustenance, especially at higher density. ATPase activities at release were significantly higher in low-density pens than in higher density pens, but development at all densities was retarded when compared with ATPase activities of fed fish.

Preliminary cost estimates for producing fish-using the rearing strategies developed in the current pen-rearing study compared favorably with the average costs of rearing salmonids in a Northwest hatchery.

INTRODUCTION

In 1983 the Bonneville Power Administration contracted the U.S. Fish and Wildlife Service to conduct a study to evaluate the feasibility of rearing and imprinting juvenile fall chinook (upriver bright) salmon (Oncorhynchus tshawytscha) in backwaters and ponds adjacent to the Columbia River. Objectives of the study are to:

- 1) Determine densities and feeding rates for rearing fish in enclosures in backwaters and ponds and to compare off-station rearing success with that in a hatchery;
- 2) Determine the optimum duration and time of release for off-station reared fish;
- 3) Rear and release fish and determine the contribution of returning adults to the Columbia River fishery;
- 4) Evaluate cost-benefit of backwater and acclimation pond rearing and develop a management plan for low-capital salmon production programs.

In 1983, after surveying all potential rearing locations from Priest Rapids Dam to John Day Dam, a backwater, Rock Creek (river km 367), and a man-made pond, Social Security Pond (river km 468) were selected as the sites best suited for the study (Novotny et al. 1984). In 1984 fish were reared at both sites at various densities and feeding rations to determine an optimum density/feeding combination for future rearing trials (Novotny et al. 1985). Additional rearing trials in

1984 evaluated the feasibility of rearing fish at low density in a backwater environment without providing supplemental food.

In 1985 fish were reared at both sites using the densities and feeding rates developed in the 1984 rearing trials. This was the first year of three years designated for rearing and releasing production level quantities of replicated, coded-wire-tagged groups. A comparison of our rearing success with traditional hatchery-based fish culture will be completed by evaluating adult returns to the rearing sites and to the fishery.

METHODS

Fish were reared at Rock Creek (RC), a backwater located 20 km upstream of John Day Dam and Social Security Pond (SSP) an artificial pond located 123 km upstream of John Day Dam, near the tailrace of McNary Dam.

Water Quality

Temperature and dissolved oxygen profiles were recorded at both rearing sites at weekly intervals from mid-March until the first week of June. Additional temperature readings were taken on a near-daily basis at a depth of one meter at both areas. Selected water quality parameters, including alkalinity, un-ionized ammonia, nitrate/nitrite nitrogen, orthophosphate, total iron, total manganese, and total organic carbon were measured prior to stocking and at release. Water quality samples were transferred to a consulting chemical laboratory for analysis.

Zooplankton

Zooplankton samples were collected weekly at SSP and RC using a Wisconsin tow net (.075 mm aperture). Sampling began when fish were stocked and continued until all fish were released from respective sites. Samples were taken from within and from outside the net pens (fed fish) at SSP and RC and from within the barrier net at RC.

Wilcoxon's paired sample test (Zar 1984) was used to test differences between densities within and outside of pens.

Fish Rearing

Eggs collected from upriver bright fall chinook salmon by the Oregon Department of Fish and Wildlife in December 1984 were transferred to Spring Creek National Fish Hatchery (SCNFH) for incubating, hatching, and initial rearing. Early broods of fry, intended for pen rearing studies, were lost in a hatchery epizootic caused by bacterial gill disease which occurred in early March. This development necessitated the use of a later brood of fry which were not available for outplanting until mid-April.

At each rearing site two groups of about 110,000 fish were stocked in twelve net pens at a density of about 430 g/m³ (at stocking) and a feeding ration of 3-4% body weight/day. Net pens were 6.1 m x 6.1 m x 2.1 m enclosing 79 m³ of rearing area and were fitted with 4.8 mm mesh netting. Fish in each group were coded-wire tagged with separate codes and transported to rearing sites during the second and fourth weeks of April at a size of 1.3-1.5 g/fish (304-342 fish/lb).

Transfer of fish to SSP and RC during the second week of April was interrupted by an increase in surface water temperatures. The increase in temperature delayed stocking of the second of two groups of 110,000 fed fish at both sites. Stocking the second groups of fish was further delayed by a reaction of the fish to medication in the hatchery. Therefore, stocking dates between replicates were separated by 11 days

at both sites, resulting in an "early" and "late" group of fish, rather than replicates. Fish were released from net pens at SSP on May 16 and 17 and at RC on May 20.

A barrier net enclosing about 0.6 hectare (volume $\sim 19,000 \text{ m}^3$ at minimum pool) was installed at RC for rearing of two groups of coded-wire tagged fish (replicates) with about 125,000 fish in each group. Fish were stocked in the barrier net on April 24 and 25 at a loading density of 16 g/m^3 ($.001 \text{ lb/ft}^3$) and at a size of 1.5 g (304/lb). Fish in the barrier net were not fed and fish predators within the enclosure at the time of installation remained throughout the rearing period. Barrier net fish were released on June 11. Numbers of fish released were estimated using a Peterson formula (Chapman version from Ricker 1975) applied to a mark-recapture exercise at the conclusion of the holding period (Appendix 1).

To evaluate the potential for transfer and rearing fish at an earlier stage of development about 9,200 fish were transported to RC at about 0.6 g/fish (760/lb) on March 25 and stocked in a small-mesh (3.2 mm) net pen; these fish were not coded-wire tagged. The fish were later divided into three groups and reared unfed in net pens at densities of 32 g/m^3 , 64 g/m^3 , and 128 g/m^3 with one replicate of each. A summarization of the schedule for the 1985 studies is included in Appendix 2.

A control group of fish for comparison of growth and physiological condition of off-station-reared fish with hatchery-reared fish was monitored at SCNFH, the natal hatchery. Two coded-wire tagged control groups of approximately 85,000 each (replicates) were released from

Little White Salmon National Fish Hatchery (LWSNFH) for return comparison with fish reared off-station (Appendix 3).

Additional information about rearing location, morphometry of the rearing sites, methodologies, and study design can be found in Novotny et al. 1984 and Novotny et al. 1985.

Length and weight measurements of fish were taken once every two weeks from stocking through release for adjustment of feeding rates, or evaluation of growth and physiological development. Gill samples were transferred to the National Marine Fisheries Service Aquaculture Field Station, Cook, Washington for determination of gill $\text{Na}^+\text{-K}^+$ ATPase activity using methods of Zaugg (1982).

Mortality of fish in pens was monitored daily by observation; carcass counts were completed when pens were crowded for collecting fish for length and weight sampling. Instantaneous growth rates ($G = \frac{\log_e W_2 - \log_e W_1}{T} \times 1000$) and condition factors ($K = \frac{W}{L^3} \times 10^7$) were determined for off-station reared fish, as well as hatchery fish, at two-week intervals. Samples of fish for routine disease checks were collected from all treatments at the mid-point of the rearing period and at release. Samples were transferred to the Lower Columbia River Fish Health Center for a health check and detection of disease (Appendix 4).

Stomachs of 5-10 fish were collected biweekly from each of the net pens and the barrier net in 1984 and 1985 to estimate percent composition and frequency of occurrence of major food items.

Groups of fish from the fed treatments at both rearing sites were

subjected to a 24-hr seawater challenge at release. An overview of the methods and results of the seawater challenge are included in Appendix 5.

One-way analyses of variance (ANOVA) were used to compare growth of early and late groups of fed fish, growth among fed fish in pens and the hatchery, and growth/ATPase activity among unfed fish in pens. A two-way ANOVA was used to analyze the results of the 24-hr seawater challenge. If significant differences were indicated by an ANOVA, a Newman-Keuls multiple range comparison (Zar 1984) or a planned comparison analysis (Sokal and Rohlf 1969) were used to compare differences among means.

Rearing costs tested thus far were compared with expenses for rearing juvenile salmon in northwest hatcheries using efficiency ratios developed by Stephen B. Mathews, PhD., U. of Washington (Senn et al. 1984). The efficiency ratios apply Present Value Theory, which has been developed by economists as a means of comparing alternative cost strategies, to compare various rearing scenarios in terms of a common denominator. The method accounts for all types of rearing scenarios, densities, and periods of use for each system. A discounting factor, which incorporates a discounting rate (the prevailing rate of interest for borrowing or lending), relates costs or benefits, which may be realized in the future, to a present-day cost estimate (for a more complete explanation please refer to pages 249-265 in Senn et al. 1984).

Accurate cost comparisons can not be made until the optimum rearing density and mean survival rate is determined after three years

of production level rearing, adult return data-are included, and expenses are modified with additional data. Therefore, these cost comparisons must be considered as preliminary and presented only to document that in general pen rearing costs are similar to hatchery rearing costs.

Adult recovery

Methods for collection of returning precocious males from Brood Year 1983 (released in May and June 1984) were evaluated in October and November. A large Lake Erie-type trap net (5 .1 cm bar mesh, 2.5 m x 46 m lead, and 2.5 m x 15 m wings) a Merwin trap net (1.6 cm bar mesh, 9 m x 46 m lead, and 9 m x 9 m wings) , gill nets (6.4 cm bar mesh), and a weir extending across the width of the stream (~ 9 m) were tested at RC. Two weirs, one extending across the release channel below SSP, and another constructed 0.4 km upstream in an adjoining channel, were used to collect returning jack salmon at SSP.

RESULTS

Water Quality

Climatic conditions in the rearing areas during 1985 increased water temperatures more rapidly than previously observed (Novotny et al. 1984, 1985). Surface temperatures rose above 15.6°C (a critical level determined in 1984 studies) at SSP and approached this temperature at RC by mid-April (Fig. 1). Air temperatures moderated within a few days, resulting in the cooling of water temperatures at both rearing areas. The warming of the pond and backwater was primarily confined to the surface layer, with underlying strata remaining below 15-16°C (Tables 1 and 2). However, water temperatures at both locations remained near 14-16°C beginning the first week of May and continued through release of fed fish. Sustained water temperatures above 15.6°C, extending to a depth of 4-6 m, were recorded from mid-May through the remainder of the periods of observation at both locations.

Dissolved oxygen concentrations at both areas remained above 10.0 mg/liter at maximum net-pen depth (approximately 2 m) throughout the study (Tables 1 and 2). Dissolved oxygen readings taken before daylight were similar to those taken at the same depths during daylight hours. Lower concentrations were observed at 6-8 m but concentrations below 6.0 mg/liter were rarely observed at either site.

Water quality conditions in the rearing areas during 1985, as indicated by seven parameters (Table 3), were similar to conditions

Figure 1. Water temperatures at Social Security Pond and Rock Creek during pen rearing studies, 1985. Stocking and release dates are indicated by arrows while the hatched line represents the critical temperature of 15.6°C.

Table 1. Temperature and dissolved oxygen profiles in the water column at Social Security Pond, 1985.

		Sample Date											
		3/14	3/22	3/29	4/3	4/11	4/19	4/24	5/1	5/9	5/13	5/17	6/4
<u>Temperature (C)</u>													
Surface		9.0	8.0	7.5	11.0	14.0	11.5	11.0	17.5	16.9	17.4	10.0	19.1
2m		6.5	8.0	7.5	11.0	13.8	11.5	11.0	14.5	14.9	14.9	17.0	19.0
4m		5.7	7.9	7.5	10.5	10.3	11.5	11.0	11.5	13.8	14.0	15.0	16.8
6m		5.0	7.8	7.0	8.5	9.7	11.5	11.0	10.5	12.2	13.4	14.0	16.5
<u>D.O. (mg/l)</u>													
Surface		15.2	13.0	14.2	14.4	13.6	10.0	11.6	12.4	13.2	11.4	12.5	10.2
2m		15.4	13.1	14.4	14.4	13.8	10.0	11.4	12.6	13.8	11.4	12.8	10.0
4m		13.6	13.2	14.4	13.9	12.0	10.0	11.6	9.2	12.0	9.2	11.9	8.2
6m		12.4	13.2	14.4	11.6	9.0	10.0	11.2	6.0	6.0	7.0	4.7	7.4

Table 2. Temperature and dissolved oxygen profiles in the water column at Rock Creek, 1985.

		Sample Date											
		3/25	4/2	4/11	4/16	4/26	5/3	5/10	5/17	5/24	5/31	6/6	6/11
<u>Temperature (C)</u>													
Surface		7.5	11.1	13.3	13.3	10.8	14.0	13.5	16.6	19.4	17.1	17.8	19.6
2m		6.0	---	---	---	10.9	12.0	13.2	15.6	18.4	15.8	17.7	17.1
4m		5.5	---	---	---	9.9	11.0	11.9	13.7	14.9	15.2	16.8	16.1
6m		5.0	---	---	---	9.4	10.0	11.6	12.6	13.9	14.6	15.7	15.9
7-8m		---	---	---	---	8.9	10.0	11.4	12.5	---	13.8	---	---
<u>D.O. (mg/l)</u>													
Surface		11.6	---	10.3	---	10.8	12.0	11.8	11.0	9.9	9.9	8.8	10.1
2m		11.4	---	10.0	---	10.7	10.2	11.1	11.3	10.3	10.0	8.6	7.5
4m		11.4	---	10.1	---	9.6	8.4	10.2	9.7	10.5	8.9	8.3	6.8
6m		11.6	---	10.6	---	8.9	8.2	8.5	7.8	6.6	7.8	6.3	6.8
7-8m		---	---	10.6	---	7.8	8.2	5.6	---	---	5.2	---	---

Table 3. Selected water quality parameters at Social Security Pond and Rock Creek during pen rearing study, 1985. (all values expressed as mg/liter).

	SSP		RC	
	4/11	5/21	4/11	5/20
Buffering capacity				
Alkalinity as CaCO ₃	89.0	63.0	53.0	73.0
Nutrient concentration				
Nitrate + nitrite as N	.330	.047	.130	.053
Orthophosphate as P	.011	.006	.033	.012
Organic carbon (total)	4.2	5.7	3.4	2.5
Products of anaerobic activity				
Iron (total)	.56	.34	.87	.18
Manganese (total)	.019	.086	.050	.028
Ammonia as N (un-ionized)	<.02	<.02	<.02	<.02

observed in 1983 and 1984 (Novotny et al. 1984, 1985). No major changes were observed in these parameters either between years or during the rearing period. Generally, both areas were adequately buffered (alkalinity > 20 mg/liter); were considered typical for moderately productive fresh water based on nutrients available for assimilation (nitrogen 0-10 mg/liter, orthophosphate .005-.020 mg/liter, organic carbon 1-30 mg/liter); and did not reflect tendencies toward high rates of anaerobic contamination or decomposition (un-ionized ammonia < .02 mg/liter; total iron < 1.0 mg/liter; total manganese < .10 mg/liter) (Boyd 1979; Krenkel and Novotny 1984; Environmental Protection Agency 1976). Iron and manganese concentrations were somewhat higher than recommended for fish culture in hatcheries (iron < 0.1 mg/liter; manganese < .01 mg/liter) (Senn et al. 1984), but were within limits recorded at various other locations within the mainstem Columbia River and its tributaries (U.S. Geological Survey 1985).

Zooplankton

Zooplankton densities were low in both rearing areas when fish were stocked but densities increased progressively over the rearing period (Fig. 2). Densities were slightly higher at SSP than at RC, but general trends during the course of the rearing period were similar at both areas. Highest zooplankton densities at both RC and SSP occurred after release of fish from pens.

There were significantly more zooplankters ($P < 0.05$) in areas outside of net pens than within them at both rearing locations (Fig.

Zooplankton- 1985

Figure 2. Zooplankton densities within (●—●) and outside of (●-----●) net pens at Social Security Pond and Rock Creek, and the barrier net at Rock Creek during pen-rearing studies, 1985. Stocking and release dates are represented by arrows.

2). While zooplankton densities tended to increase outside of the pens, densities within the pens remained the same or decreased. Densities of zooplankton within the barrier net were similar to those taken outside of the net during the first three weeks of the rearing period. However, zooplankton numbers were highest (about 200/liter) in the barrier net during the final 2-3 weeks of rearing.

Mean numbers of zooplankton for the entire rearing period were higher outside than inside of pens at both locations. Large-sized organisms (cyclopoids, calanoids, and daphnids) were less abundant within pens containing fish which were fed while smaller-sized zooplankters were found in similar numbers within, and outside of these pens (Table 4).

Fish Rearing at Off-Station Sites

Fish reared in pens and fed

Fish were stocked in net pens at SSP and RC at a mean size of 1.3 g/fish (342 fish/lb) on April 11 and 12, respectively, and at a size of 1.5 g (304/lb) on April 22 and 23, respectively. Fish were fed throughout the study at the regular hatchery rate of 3-4% body weight/day. Number of fish stocked/pen averaged about 19,200 at both locations with mean loading densities of 321 g/m^3 ($.020 \text{ lb/ft}^3$) for early groups stocked and 353 g/m^3 ($.022 \text{ lb/ft}^3$) for the late groups (Table 5). Numbers of fish stocked/pen were slightly higher than stocked at high density during 1984 rearing trials ($\bar{X} = 17,350/\text{pen}$) to compensate for the smaller-sized fish available for stocking in 1985

Table 4. Taxonomic composition of zooplankton (mean number/liter) taken from within and from outside net pens at Social Security Pond and Rock Creek, and a barrier net at Rock Creek, 1985.

Taxon	SSP		RC		RC (Barrier net)	
	out	In	out	In	out	In
Cyclopoida	21.9	4.2	12.9	0.6	63.6	33.9
Calanoida	0.5	0	0.1	0	0.3	0
<u>Daphnia</u>	9.0	1.0	0.3	0.2	24.9	5.2
<u>Bosmina</u>	4.5	4.4	0	0.8	280.8	114.5
<u>Chydorus</u>	0.5	1.6	0	0	0.5	0.2
Other Cladocera ^a	0.2	0.6	0	0	1.2	0.7

^aIncludes Alona, Diaphanosoma, Ceriodaphnia, and Polyphemus.

Table 5. Summary of stocking numbers and densities, mortalities, and numbers, sizes and weights at release in early and late groups of fed fish at Social Security Pond and Rock Creek, 1985. (Numbers represent mean values for early and late groups, respectively.)

	<u>Early Group</u>		<u>Late Group</u>		
	<u>SSP</u>	<u>RC</u>	<u>SSP</u>	<u>RC</u>	<u>Mean</u>
Stocking summaries:					
Number/pen	18,042	19,910	18,657	19,565	19,244
Weight/pen (lb/ft ³)	.020	.021	.022	.023	.022
Weight/pen (g/m ³)	321	337	353	369	345
Mortalities/Pen:					
Natural (%)	205	198	98	60	140
	(1.1%)	(1.1%)	(0.5%)	(0.3%)	(0.8%)
Sample	186	209	134	151	170
Release summaries:					
Number/pen	18,451	19,503	18,425	19,354	18,933
Size (g)	4.6	4.7	4.0	3.3	4.2
Size (fish/lb)	99	96	113	139	112
Weight/pen (lb/ft ³)	.067	.072	.058	.050	.062
Weight/pen (g/m ³)	1,075	1,155	930	802	990

(1.3-1.5 g/fish versus 2.0 g/fish in 1984). However, initial loading densities used in 1985 (weight/unit volume rearing area) were lower than used in 1984--433 g/m³ (.027 lb/ft³).

Growth over the entire rearing period was higher for fish stocked during the second week of April (early fish) than for control fish in the hatchery or for fish stocked during the last week of April (late fish) (Fig. 3). At release, size of the late group at SSP was similar to size of fish in the hatchery (not significant $P > 0.051$, but at RC size of the late group at release was significantly smaller than in the hatchery ($P < 0.05$; Appendices 6 and 7).

Mean size of fish at release in the early group was 4.6 g (99/lb) at SSP and 4.7 g (96/lb) at RC; late groups averaged 4.0 g (113/lb) at SSP and 3.3 g (139/lb) at RC (Table 5). Size of early fish was significantly larger than the late groups at both sites ($P < 0.05$; Appendix 8). Mean release size of fed fish from all pens at SSP was 4.5 g (101/lb) and at RC, 3.8 g (118/lb). Weight of fish released/pen ranged from 802 to 1155 g/m³ (.050 to .072 lb/ft³) for early and late fish at both locations.

Instantaneous growth rates were highest during the first two-three weeks after stocking (Table 6). Growth rates of the early group of fed fish slowed considerably during the final two weeks of the rearing period. Fed fish in pens all had higher growth rates than hatchery fish, or fish in other enclosures.

Condition factors (K) of fed fish were higher than those reared in any of the other enclosures, especially during the initial rearing period. Fish of the early groups had slightly higher K-factors than

Figure 3. Growth of early and late groups of fall chinook juveniles in pens at Social Security Pond and Rock Creek, and of the same brood of fish held in the hatchery as a control; and growth of fish reared in a barrier net at RC in relation to growth of hatchery controls.

Table 6. Summary of instantaneous growth rates and condition factors for fish reared at Social Security Pond, Rock Creek, and Spring Creek National Fish Hatchery, 1985.

	<u>Instantaneous growth</u>			<u>Condition factor</u>		
Social Security Pond						
Fed fish:	<u>4/25</u>	<u>5/6</u>	<u>5/16</u>	<u>4/25</u>	<u>5/6</u>	<u>5/16</u>
Early fish	50	43	17	146	133	128
Late fish		42	37		136	128
Rock Creek						
Fed fish:	<u>4/26</u>	<u>5/7</u>	<u>5/20</u>	<u>4/26</u>	<u>5/7</u>	<u>5/20</u>
Early fish	46	49	14	152	143	129
Late fish		23	34		130	128
Barrier net:	<u>5/2</u>	<u>5/22</u>	<u>6/11</u>	<u>5/2</u>	<u>5/22</u>	<u>6/11</u>
	-11	23	2	113	119	112
Unfed fish:	<u>5/11</u>	<u>5/23</u>	<u>6/5^a</u>	<u>5/11</u>	<u>5/23</u>	<u>6/5</u>
Density 1 (32 g/m ³)	12	-1	52	119	111	137
Density 2 (64 g/m ³)	7	1	51	110	108	136
Density 3 (128 g/m ³)	10	-13	49	112	102	135
Hatchery fish: (Spring Creek NFH)	<u>4/17</u>	<u>5/8</u>	<u>6/11</u>	<u>4/17</u>	<u>5/8</u>	<u>6/11</u>
	11	38	14	132	110	100

^a After 6 days of feeding.

did fish in the late group after the initial stages of rearing. Condition factors declined somewhat in all groups of fish over the rearing period.

Mortality was low in all pens in 1985 (Table 5) with a cumulative mortality of 1.1% for fish stocked early at both locations, and 0.5% and 0.3% for late groups stocked at SSP and RC, respectively. Mortality was high only during the initial two weeks after stocking of early fish at SSP and RC (Fig. 4; Appendix 9). The initially high mortalities may have resulted from a combination of transport stress and the relatively warm water temperatures present at SSP and RC during stocking, especially at SSP. Fish in both locations were fed prophylactic treatments of TM-50 medicated (3-4 g teramycin/45 kg fish) Abernathy Dry Diet when it appeared extended water temperatures might exceed 15.6°C. Even though water temperatures were commonly near, or slightly below temperatures where heavy mortalities were observed during the 1984 trials, losses were minimal. No disease, or mortality due to disease, was identified in any of the fed fish after stocking (Appendix 4).

Gill Na⁺-K⁺ ATPase activity of fish reared at both locations increased progressively from stocking through release (Fig. 5). ATPase levels were slightly higher in early fish than in late fish on all occasions. Activity increased at a faster rate in fish reared at both areas than in hatchery fish and at release ATPase levels were significantly higher than levels observed in the hatchery ($P < 0.05$). However, during the initial 2-3 weeks after stocking, ATPase levels were similar to those observed in the hatchery. ATPase levels rose

Figure 4. Weekly mortality of early (o—o) and late (●—●) groups of fall chinook juveniles reared in pens in relation to critical temperatures (15.6°C) at Social Security Pond and Rock Creek 1985. (Dashed line represents unacceptably high mortality of 0.1 %/day.)

Figure 5. ATPase activities (micromoles Pi/mg P/hour) of fall chinook juveniles in early and late groups at Social Security Pond and Rock Creek, in the barrier net and unfed fish (at release), and in hatchery controls. The dashed line represents a target ATPase activity above which fish are assumed to be in a high state of smoltification (W. Zauq, NMPS, Cook, WA, personal communication).

above 20 micromoles Pi/mg prot/hr, considered indicative of smoltification activity in salmonids (W. Zaugg, personal communication), in both groups of fish at SSP by release, but remained below this level for fish at RC.

Results of a 24-hr seawater challenge test (Appendix 5) were generally in agreement with gill ATPase analyses, i.e. fish reared off-station were undergoing physiological changes indicative of the onset of smoltification, whereas those in the hatchery exhibited a delay in smoltification activity. At release, fish from SSP and RC were able to regulate blood-Na⁺ levels below those of fish in the hatchery (Appendix 5). However, the differences between blood-Na⁺ levels in off-station fish and the hatchery control group were significantly different only for the RC fish (ANOVA, $P < 0.05$).

Fish reared in pens without feeding

Densities of fish reared in pens without supplemental feeding were two, four, and eight times higher than examined in 1984, (Novotny et al. 1985) resulting in stocking of slightly over 1000, 2000, and 4000 fish/pen (Table 7). No weight gain was observed after four weeks of rearing, and very little growth in length occurred in any of the densities tested (Fig. 6). Mortalities increased during the fourth and fifth weeks of rearing, necessitating the prophylactic feeding of fish according to dosages recommended by USFWS fish health personnel (3-4% body weight/day with TM-50) over the final 6-7 days of rearing. These fish were required for a scheduled release within the barrier net as the "marked" portion of a subsequent mark/recapture estimate. The

Table 7. Total number of fish stocked per pen, mortality, release size (number/lb) and ATPase activity for unfed fish reared in pens at densities of 32, 64, and 128 g/m³ (.002, .004, and .008 lb/ft³), Rock Creek, 1985.

Characteristic	Density					
	32 g/m ³		64 g/m ³		128 g/m ³	
	A	B	A	B	A	B
Number fish stocked/pen	1056	1016	2071	2001	4392	4106
Mortality (sample mortality)	21 (160)	30 (2.5%) (160)	87 (160)	313 (9.8%) (190)	1109 (173)	821 (22.7%) (186)
Release weight (fish/lb)	116	129	126	152	158	156
ATPase activity (micromoles (Pi/mg prot/hr)	16.2	13.3	15.9	10.9	9.7	12.0

Figure 6. Weights (g) and fork lengths (mm) of unfed fish in pens at Rock Creek reared at low (.002 lb/ft³), intermediate (.004 lb/ft³) and high (.008 lb/ft³) densities, 1985.

increased mortality was not an epizootic, but the apparent lack of adequate food for growth. Presence of enteric redmouth in one of the high density pens (Appendix 4) did present potential conditions for an epizootic as water temperatures exceeded 15-16°C in the water column within the pens.

Feeding of these previously unfed fish resulted in a nearly doubling of the mean weight/fish and an increase in mean length of 7-9 mm/fish for all densities tested (Fig. 6). At release the largest fish (3.7 g-1 22/lb, mean size), were in pens with the lowest density, the smallest fish (2.9 g-157/lb, mean size), in pens with the high density and intermediate sized fish (3.3 g-1 39/lb, mean size) in pens with the medium density (Table 7). Fork lengths of fish at low and medium densities were not significantly different ($P > 0.05$), but fish at both densities were significantly larger than fish in high density pens ($P < 0.05$; Appendix 10). Total mortality of fish over the entire rearing period was 2.5% for those reared at low density, 9.8% at the intermediate density, and 22.7% at the high density.

Instantaneous growth of fish not fed during the first four weeks of rearing was low in relation to that observed in fish which were fed but similar to the low rate of growth observed by fish in the barrier net (Table 6). Some growth took place during the initial two weeks of rearing, but during the second two-week period no growth was observed for fish reared at any of the densities tested. Fish exhibited large growth increases after being fed for 6 days.

Slight differences were noted in condition factors among fish reared at the three densities, although fish reared at low density

tended to have a slightly higher condition factor than those reared at higher densities throughout the rearing period. After feeding of fish condition factors increased to levels observed in those fish which were fed and reared in higher density pens.

ATPase activity of unfed fish at release ranged from 9.7 to 16.2 micromoles Pi/mg prot/hr with highest levels occurring at lowest density and lowest levels at high density (Table 7). At release, activities were significantly higher at low density than at high density ($P < 0.05$) but levels of fish in medium density pens were not different than either of the other densities ($P > 0.05$; Appendix 10).

Fish reared in barrier net

Fish in the barrier net did not grow in weight during the first two weeks after stocking; after two weeks mean weight/fish was 0.2-0.3 g less than the 1.5 g at stocking (Fig. 3). After seven weeks of rearing (April 24 - June 11) mean size of fish in the barrier net was 2.7 g (169/lb). Fish in the hatchery grew considerably faster and their mean size was nearly 2.0 g larger at release than those in the barrier net.

A negative index of instantaneous growth was observed in barrier net fish during the initial two weeks of rearing, and while growth rates did subsequently increase, they were low in relation to those observed in fish which were fed (Table 6). Condition factors were relatively low in the barrier net fish throughout the rearing period.

Mortality of fish in the barrier net was estimated to be 49% over the entire rearing period. Estimates were determined from the

difference in number of fish stocked (254,194), and the estimated number remaining (129,764 - mark/recapture) within the enclosure at the conclusion of the rearing period (Appendix 1). No disease was detected in fish reared in the barrier net and routine health monitoring of fish did not reveal a health-related situation which could have resulted in the estimated high mortality. Fish predators were not removed from the enclosure, and predatory birds were commonly observed perched on the perimeter of the enclosure. Therefore, it was assumed that predation was the primary cause of the mortality in the barrier net. Estimates of taxonomic composition and size distribution of the 153 predators collected at the end of rearing was similar to that observed in 1984 (Appendix 11).

Gill ATPase activity of barrier-net fish was low at stocking and remained low throughout rearing, with no indication of the onset of smoltification (Fig. 5). Activity levels were below those of the hatchery controls and for any of the other groups of fish reared off-station in 1985.

good habits of unfed fish in barrier net and pens, 1984 and 1985

Zooplankton comprised the major portion of food items found in stomachs of fish reared in the barrier net in 1984 and 1985 and in pens in 1984 (Table 8). In 1984 zooplankton comprised more than 99% of the diet (by number) of fish in the barrier net and pens, with Daphnia accounting for about 75% and Cyclopoida about 17% of the total. Zooplankton percent was slightly lower in the diet of fish in the barrier net in 1985 (82%). In 1985, Daphnia accounted for a much lower

Table 8. Preliminary summary of food items identified in stomachs of fall chinook salmon during rearing in the barrier net, 1984 and 1985, and in unfed pens, 1984, at Rock Creek. (F.O.--frequency of occurrence; %N--percent of total by number.)

Food item	Barrier net (1984)		Barrier net (1985)		Unfed pens (1984)	
	F.O.	%N	F.O.	%N	F.O.	%N
Abernathy dry	--	--	6.0	--	--	--
Zooplankton		99.9		81.7		99.5
Cyclopoida	(93.3)	(17.5)	(64.0)	(39.2)	(93.3)	(16.5)
Calanoida	--	--	(2.0)	(T)	--	--
Daphnia	(93.3)	(73.3)	(30.0)	(7.6)	(93.3)	(80.6)
Bosmina	(30.0)	(7.1)	(56.0)	(33.6)	(13.3)	(2.1)
Other ^a		(2.0)		(1.3)		(0.3)
Insects				4.5		0.4
Chironomidae	(3.3)	(TT)	(20.0)	(2.7)	(6.7)	(0.1)
Odonata	(3.3)	(T)	(2.0)	(0.1)	--	--
Trichoptera	--	--	--	--	(3.3)	(0.1)
Hymenoptera	--	--	(4.0)	(0.2)	--	--
Diptera (other than Chironomidae)	--	--	(2.0)	(0.7)	(6.7)	(0.2)
Other ^b		(T)		(0.8)		(T)
Arachnida	3.3	T	10.0	1.9	3.3	0.1
Nematoda	--	--	--	--	3.3	0.1
Bryozoa	--	--	18.0	11.3	3.3	T
Fish parts	3.3	T	--	--	--	--
Inedibles ^c	3.3	T	12.0	0.3	3.3	T

^aMiscellaneous zooplankton including Leptodora, Diaphanosoma, Chydorus, Alona, Ceriodaphnia, and unidentifiable parts.

^bMiscellaneous insects including Isoptera, Thysanoptera, Coleoptera, and unidentified parts.

^cRocks and detritus.

percent of the total number of food items (7.6%), but Cyclopoida (39.2%) and Bosmina (33.6%) comprised a larger portion of food items. Insects were not a common food item found during either year, or in either enclosure type.

The only other item identified which comprised a relatively large portion of the total number of food components was bryozoan statoblasts, which were common but because of their small size were considered an incidental contribution to the total diet.

Preliminary observations of the barrier net fish stomachs from 1985 indicated that the fish did not feed during the first two weeks after stocking. Subsequent stomach analyses revealed that the fish took progressively more food items over the remainder of the rearing period, and at release, most stomachs were full of zooplankton.

Rearing and Release of Hatchery Fish

Growth and size of fish were similar over the period and ATPase activities remained low in fish at both hatcheries; activities did not reach 20 micromoles Pi/mg prot/hr at either site by the release date (Appendix 12). Sizes of fish at release were 4.5 g (102/lb) at SCNFH (on June 12) and 5.2 g (88/lb) at LWSNFH (on June 19).

Instantaneous growth of hatchery fish was moderate in relation to fed fish in pens, but generally higher than in other enclosures (Table 6). Condition factors of hatchery fish were lower than the higher estimates for fed fish in pens and tended to decrease over the period of observation.

Cost Estimates for Rearing Fish

Efficiency ratios (Senn et al. 1984) using rearing scenarios tested in the pen rearing study thus far were compared with each other, with calculated efficiency ratios for net-pen rearing at higher density, and with the rearing of fish in a concrete hatchery raceway (Appendix 13 & Table 9). Costs were estimated from actual expenses incurred during 1984 or 1985 and standardized for rearing and production of 1000 lb of fish.

The most costly method (lowest efficiency ratio) of the methods compared was the barrier net, but fish reared at low density in net pens were also relatively expensive to produce (Table 9). Increased density in high-density pens resulted in a higher efficiency ratio and lowered expenses; this was the least costly method of the scenarios tested in the present study. Higher densities in net pens would produce an additional lowering of costs, as indicated using the rearing density of .300 lb/ft³, which has been used successfully in other net pen culture operations (Senn et al.). In all of the above rearing scenarios, increased labor and facilities costs were the primary causes of the increased expense for rearing like numbers of fish at lower densities. Plumbing and water were not a factor in net pen or barrier net rearing; additionally, food costs (unfed fish) and labor were not an expense in barrier net rearing. Efficiency ratios calculated for a concrete raceway were higher than those calculated for low density net-pen or barrier net culture, but Lower than efficiency ratios in net-pen culture at the high density tested in current pen rearing

Table 9. Rearing cost summaries and calculated efficiency ratios for rearing 1000 lb of fish in net pens at densities tested in 1984 and densities suggested as "rearable" by Senn et al. 1984; in a barrier net using the scenario tested in 1984 and 1985; and in a concrete raceway using cost estimates developed by Senn et al. (1984) and actual hatchery expense.

Method	Pond/facility ^a	Plumbing	Water	Food ^b	O & M ^c	Labor ^d	Efficiency	Cost
							ratio	
							lb/\$	\$/lb
Net pen-low density (.043 lb/ft ³)	25032	0	0	94	1252	2417	.14	7.14
Net pen-high density (.086 lb/ft ³)	12618	0	0	190	631	1218	.26	3.85
Net pen-(.300lb/ft ³)	3605	0	0	300	180	348	.33	1.37
Barrier net (.001 lb/ft ³)	35914 ^e	0	0	0	1796	0	.13	7.69
Concrete raceway ^f	6440	6000	631	600	2350	835	.18	5.56

a/ Net pen costs based on an expected useage life of 20yr for frames, feeders, and floats and 7yr for netting (Senn et al. 1984).

b/ Food costs based on the use of Abernathy Dry at \$.45/lb for net pens and the use of OMP at \$.60/lb for hatchery.

c/ Operation and maintenance expense figured at 5% of capital investment/year for net pens and barrier net, and at \$2.35/lb produced at the hatchery (J. Bodle, Complex Manager, Little White Salmon National Fish Hatchery).

d/ Labor expense figured on basis of pay of \$25,000/year/man. Net pens required .012 man-year/pen and hatchery raceways required .033 man-year/1000 lb of fish produced,

e/, Barrier net expenses include an expected mortality rate of 40% (average of 1984 & 1985), assuming predators are not removed from within the enclosure. Expected useage life of barrier net was figured at 10yr.

f/ Hatchery costs based on an expected useage life of 50yr for the raceway and 25yr for plumbing.

Table 10. Summary of jack salmon captures and time expended using the various methods at Rock Creek with gill nets, a Merwin trapnet, and a Lake Erie trapnet, fall, 1985 (each month was divided into four weeks, but there was overlap in fishing periods using the Merwin trapnet).

	SEPT				OCT				NOV				Total
	1	2	3	4	1	2	3	4	1	2	3	4	
Gill Net													
Hours fished	0	4	2	3	2	20	47	6	0	0	0	0	84
Number jacks	0	0	0	0	0	0	14	0	0	0	0	0	14
Merwin Trap													
Days fished	0	0	0	2	9	11	6	9	7	8	7	5	64
Number jacks	0	0	0	0	0	4	1	7	26	30	11	0	79
Lake Erie Trap													
Days fished	0	0	0	0	0	0	4	0	0	0	0	0	4
Number jacks	0	0	0	0	0	0	2	0	0	0	0	0	2
													Total jacks recovered 95

studies or in the .300 lb/ft³ density estimates used for comparison. Hatchery expenses included plumbing and water costs as well as expenses incurred using the other rearing scenarios.

Adult Recovery - 1985

Jack salmon were collected effectively by both trap and gill nets at RC (Table 10). A total of 95 fish were captured using all methods at RC. Large mesh netting in the Lake Erie trap net (5.1 cm bar) tended to gill fish; the Merwin trap net with smaller mesh netting (1.6 cm) appeared to be equally effective and fish were not gilled in the netting. Gill nets (6.4 cm bar) also captured jacks effectively, but use was discontinued after several adult steelhead (Salmo gairdneri) were captured.

Weirs constructed at both sites were least effective in capturing jack salmon. At RC flows were marginal throughout the fall months, making it difficult for fish to swim into the weir. At SSP flows were adequate in both weir locations, but only four jacks were recovered. No other methods were tested at SSP.

DISCUSSION

A planned stocking date for the first week of April for transferring upriver bright fall chinook salmon from SCNFH to SSP and RC was delayed by the loss of the fish at the hatchery during an epizootic of bacterial gill disease in March; high water temperatures further delayed and complicated the transfer of fish from the hatchery. The net results of these events were the shortening of the potential period of rearing from 8-10 weeks to 3-8 weeks and the release of fish at a smaller size and earlier stage of smolt development than desired.

Even though temperatures and environmental conditions at off-station sites resulted in growth rates and smolt development exceeding that in the hatchery control fish, the reduced length of rearing allowed less time for growth and smolt development. Thus the reduced length of rearing was disadvantageous for the off-station rearing strategy.

Fish were released at a similar size as fall chinook released during normal hatchery operation (Vreeland 1985) but they were smaller than the desired release size of about 5.09 g (90/lb). Temperature increases above 15-16°C in 1984 rearing trials were associated with increased rates of mortality due to disease. Therefore, extending the rearing period of fed fish in pens in order for them to reach a larger size could have resulted in unacceptably high mortality rates. Consequently, early and late groups of fed fish in pens at SSP and RC were somewhat undersize at release, but fish were healthy and condition factors were good in relation to hatchery fish or fish in other

enclosures. Smolt development of fed fish (as indicated by rising ATPase activities) proceeded as expected, based on the results of the 1984 rearing trials.

Densities at which fish can be reared unfed and still maintain acceptable growth appear to be lower than those used in unfed treatments in 1985. The growth of unfed fish was dependent on their primary food source, zooplankton. Thus, the magnitude and occurrence of earlier or later increases in zooplankton densities probably influenced the growth rates and development of unfed fish. Since growth of unfed treatments in 1984 was considered very good, either the lower density used in those treatments ($.001 \text{ lb/ft}^3$) was close to an optimum, or zooplankton populations were more nearly adequate. Growth was retarded in unfed treatments, but the fish responded well once fed, and physiological change did occur among the various densities.

Fish reared in the barrier net did not grow as well in 1985 as in 1984, and growth and physiological development were slower in 1985 than for fish reared in the hatchery.

Zooplankton densities were relatively low when fish were stocked in the barrier net, and may have been lower than required for sustenance during the transition period when the fish were forced to adjust from a hatchery regimen to reliance on the natural food base. In addition, the fish stocked in the barrier net were apparently overmedicated a few days prior to transfer from the hatchery (S. Leek, personal communication, Lower Columbia River Fish Health Center). Overmedication appeared to have temporarily sickened the fish, imposing additional stresses and retarding feeding once they were transferred

into the barrier net. The poor growth observed in fish in the barrier net, especially during the initial weeks of rearing, was probably the result of the combined effects of low zooplankton densities and their condition when stocked.

Mortality of fish in the barrier net (49%) was higher in relation to that observed for fish reared under similar conditions in 1984 (30%) or in any of the other groups reared. Disease was not detected in barrier net fish, even after surface waters warmed above 16°C. Therefore, the high mortality was apparently due to predation since fish predators were not removed from within the barrier net and birds were commonly observed around the perimeter of the enclosure. Other causes of mortality during the initial two weeks may have been due to the condition of the fish at transfer and the low zooplankton densities during that period.

The groups of fed fish which were stocked late at SSP and RC, were from the same group of hatchery fish as used to stock the barrier net. While these fish at SSP appeared to grow well over the period of rearing, growth of those at RC was retarded, especially during the first two weeks. However, neither of the late groups of fed fish underwent a period of no growth similar to that observed in the barrier net. Apparently, the SSP fish were either not affected by medication in the hatchery or they were quicker to respond once transferred to the off-station location.

Off-station methods tested during the pen-rearing trials, especially at higher density, are comparable or slightly lower than costs of rearing fish in hatcheries. The net-pen enclosures require

relatively high labor demands and initial capital investment, especially at low rearing density. Higher densities in the net pens as well as the barrier would decrease costs and improve efficiency.

Additional rearing scenarios including rearing of fish at higher densities in net pens and removal of predators from the barrier net will be incorporated into the 1986 studies to test the effects of higher densities and reduced predation on efficiency ratios. The health and quality of these fish, as well as those being reared at regular densities according to the 1985 study plan, will be closely monitored by expanding physiological observations to include blood plasma thyroxin and cortisol analyses. The length of the rearing period, especially in net pens, is limited by water temperatures. Therefore, to increase production by lengthening the growth period and to grow the fish to a desirable size (>90/lb), fish will be transferred from the hatchery to off-station sites at a very early stage of development ($\approx$ 800-900/lb), which may increase the period of off-station rearing by 4-6 weeks.

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Appendix 1. Barrier net calculations for determination of mortality and numbers of fish in area at release, 1985.

Summary:

4/24 & 4/25/85 254,194 fish stocked in barrier net

6/4 & 6/5/85 29,902 "marked" fish^a introduced into barrier net

6/10 & 6/11/85 barrier net sampled with 100-foot seine

(1) 2328 unmarked and 679 marked

(2) 1022 unmarked and 270 marked

Calculations:

- a. 254,194 fish in original stocking group including 7675 (3.0%) with marks. Based on mortality observed during 1984 rearing trials, 30% of the original marks were assumed mortality losses for mark-recapture purposes.^b Therefore, 5,372 fish with marks were included from the original stocking group in the 'marked' group released into the barrier net on 6/4 and 6/5.

- b. Peterson formula (Chapman version) from Ricker (1975):

$$N = \frac{(M+1)(C+1)}{(R+1)}$$

M = 29,902 + 5372 total number of "marked" fish in barrier net

C = 4299 total fish in seine samples

R = 949 total "marked" fish in seine samples

c. $N = \frac{(35,274 + 1)(4299 + 1)}{(949 + 1)} = \underline{159,666}$ fish in barrier net on 6/11

- d. 159,666 - 29,902 (stocked on 6/5) = 129,764 total number of originally stocked group released from barrier net on 6/11/85

- e. 254,194 - 129,764 = 124,430 or 49% mortality

^a"Marked" fish in these calculations refer to fish introduced into the barrier net with no adipose fin clip (about 3% of the original group stocked remained with adipose fins after coded wire tagging).

^bUsing a 50% scenario increased mortality estimates by about 3%.

Appendix 2. Chronological schedule of events for upriver bright chinook salmon rearing, transfer, and release for 1985 pen-rearing studies.

9/1/84 - 9/30/84	Adults captured in Bonneville Hatchery (Oregon Dept. of Fish and Wildlife) fish traps and fish ladder at Bonneville Dam.
12/1/84 - 12/22/84	Eggs taken at Bonneville Hatchery and transferred to Spring Creek and Little White Salmon National Fish Hatcheries.
1/1/85 - 3/1/85	Fish reared and ponded at the hatcheries.
3/25/85	Fish (9196) transferred at 760 fish/lb from SCNFH to smallmesh pen (1/8") at RC.
4/11/85	Fish stocked in pens no. 7-12 at SSP (111,943) - tag code H-5-7-2.
4/12/85	Fish stocked in pens no. 1-6 at RC (119,457) - tag code H-5-7-1.
4/22/85	Fish stocked in pens no. 1-6 at SSP (113,057) - tag code H-5-7-3.
4/23/85	Fish stocked in pens no. 7-12 at RC (117,388) - tag code H-5-7-4.
4/2,/85	Fish in small-mesh pen split into 7 pens (unfed fish).
4/24/85	Fish stocked in barrier net (123,450) - tag code H-5-7-5.
4/25/85	Fish stocked in barrier net (130,744) - tag code H-5-7-6.
5/16/85 - 5/17/85	Fish released at SSP.
5/20/85	Fish released from pens no. 1-12 at RC.
6/5/85	Unfed fish from low-density pens transferred to barrier net. An additional unmarked number of fish (18,663) transferred from the hatchery to barrier net for mark-recapture population estimate.
6/11/85	Barrier net fish released.
6/11/85	Control fish at Spring Creek Hatchery transferred to mid-Columbia basin for release.

6/20/85 Little White Hatchery control (approximately 170,000)
released at hatchery.

Appendix 3. Tagging and release summaries for fish coded-wire tagged (CWT) and released from net pens at Social Security Pond and Rock Creek, a barrier net at Rock Creek, and from the Little White Salmon National Fish Hatchery, 1985.

	<u>Social Security Pond</u>		<u>Rock Creek</u>		<u>Barrier Net</u>			
Stocking data	4/11/85	4/22/85	4/12/85	4/23/85	4/24/85	4/25/85		
Code	H-5-7-2	H-5-7-3	H-5-7-1	H-5-7-4	H-5-7-5	H-5-7-6		
Total stocked	111,943	113,057	119,457	117,388	123,450	130,744		
Total mortality	<u>2,352</u>	<u>1,397</u>	<u>2,432</u>	<u>1,451</u>	<u>60,490</u>	<u>64,065</u>		
Total released	109,591	111,660	117,025	115,937	62,960	66,679		
Ad. clip/no CWT	2,411	1,932	2,185	3,305	916	967		
No clip/with CWT	4,219	643	6,798	1,271	--	--		
No clip/no CWT	<u>3,792</u>	<u>3,679</u>	<u>11,897</u>	<u>11,441</u>	<u>1,889</u>	<u>2,151</u>		
Total marked CWT released	99,169	105,406	96,145	99,919	60,155	63,561		
	<u>Hatchery Control (A)</u>				<u>Hatchery Control (B)</u>			
Stocking date	- 6/20/85 -				- 6/20/85 -			
Code	5-12-50	5-12-51	5-12-56	5-12-57	5-12-52	5-12-53	5-12-54	5-12-55
Total stocked	22,393	23,100	21,864	26,499	20,075	21,158	25,467	25,505
Ad. clip/no CWT	<u>3,645</u>	<u>3,760</u>	<u>840</u>	<u>1,019</u>	<u>3,268</u>	<u>3,444</u>	<u>979</u>	<u>980</u>
Total CWT/group	<u>19,455</u>	<u>19,340</u>	<u>21,024</u>	<u>25,430</u>	<u>16,807</u>	<u>17,714</u>	<u>24,488</u>	<u>24,525</u>
Total CWT/replicate	85,249				83,534			

Appendix 4. Summary of health inspections completed for off-station reared fish by the Lower Columbia River Fish Health Center, cook, WA., 1985.

Date	Summary finding
5/10/85	Five fish recovered from fed pens at RC which appeared to be suffering some form of stress - no pathogens detected.
5/16/85	Ten fish from each pen at SSP examined for viruses, <u>Myxosoma</u> , and bacteria - no pathogens detected. ^a
5/20/85	Ten fish/pen from fed fish in pens at RC examined for viruses, <u>Myxosoma</u> , and bacteria - no pathogens detected. ^a
5/28/85	Enteric redmouth disease (ERM) isolated from one of the two pens with high densities of unfed fish; ERM was not found in other pens.
6/11/85	Harrier net fish examined at release for virus, <u>Myxosoma</u> , and bacteria - no pathogens detected.

^aFlexibacter columnaris was present in fish but was not causing losses at release time.

Note: Prior to transfer from Spring Creek National Fish Hatchery all fish used for stocking off-station rearing enclosures were included in routine health checks completed by the Lower Columbia River Fish Health Center. No disease was detected in any of the fed fish reared at Social Security Pond or Rock Creek.

Appendix 5.

SEAWATER CHALLENGE SUMMARY - 1985

Introduction

A seawater (SW) challenge test was developed by Clark and Blackburn (1978) to test hypo-osmoregulatory ability of juvenile salmon when transferred from fresh water (M) directly into SW. Analysis of blood-plasma Na^+ levels of fish can be used to predict readiness of juveniles for migration and adaptability to SW, i.e. smoltification. The ability of fish to regulate blood-plasma Na^+ levels after exposure to SW is indicative of a high hypo-osmoregulatory ability and is characteristic of fish which have smolted. However, elevated plasma- Na^+ levels are characteristic of fish which have not undergone the smoltification process (Parr).

Analyses of plasma- Na^+ levels of fish transferred from FW directly into SW have indicated that highest Na^+ levels occur 24 hr after SW entry; hence, most SW challenge tests involve a 24-hr SW exposure (Clarke and Blackburn, 1978). Some investigators have suggested that an absolute plasma- Na^+ level after 24-hr SW exposure be used as a criterion for separating smolts from parr (Clarke and Blackburn, 1978). However, because plasma- Na^+ levels appear to be dependent on several factors, including species and rearing location, it may be best to consider relative differences between control and treated groups of fish exposed to SW than to assign an absolute 24-hr Na^+ level as the criterion for smolt status of individual groups of fish (Dickoff et al. 1985).

Appendix 5 (continued)

The goal of the SW challenge tests in the-present study was to assess the degree of smoltification of fish reared at and released from Social Security Pond (SSP) and Rock Creek (RC) and to compare the results with those from the same brood stock of fish reared at Spring Creek National Fish Hatchery (SCNFH). Seawater challenge results were also compared with gill Na⁺-K⁺ ATPase activities of the test groups.

Methods

Age-0 fall chinook salmon from rearing pens (fed fish) at SSP and RC and from the control group held at SCNFH were tested in a 24-h SW challenge 3-5 weeks after transfer of fish to off-station rearing sites. Two groups of fish were tested from each site: those reared off-station for approximately five weeks (Group 1) and those reared off-station for about three weeks (Group 2). Fish from SSP and RC were tested in separate SW challenge tests. Each test included one FW control group and one replicated SW group for each area (SSP or RC; SCNFH).

Fish were collected at off-station rearing sites and transported to wet-lab facilities at the Willard Field Station (SNFRC). Fish were transported in cooled, aerated containers of 10 mg/l MS 222 at a density of about 1.0 fish/liter. On arrival, fish were acclimated in FW (SSP, RC: 15°C; SCNFH: 10°C) for 17-26 hours before direct transfer into full strength SW (30 ‰). Following a 24-h SW exposure, tails were severed and blood samples taken for plasma-Na⁺ determinations. Due to the small size of the test fish ($\bar{x}$ wt = 3.3 g), blood from five

Appendix 5 (continued)

fish was pooled to obtain an adequate volume of plasma for each sample.

Plasma-Na⁺ levels were determined utilizing a Corning Model 480 flame photometer following methods developed by the manufacturer (Corning 1983). In addition, plasma of a known Na⁺ level was run after approximately every 15 samples as a quality control. Gill Na⁺-K⁺ ATPase analyses were completed by the National Marine Fisheries Service, Cook, WA, using methods developed by Zaugg (1982). Data were analyzed using a two-way analysis of variance and a Newman-Keuls (N-K) multiple-comparison test.

Results and Discussion

Social Security Pond

In each group tested, plasma-Na⁺ levels of fish exposed to SW were significantly higher than corresponding levels of FW control fish ($P < 0.05$; Appendix Fig. 1). After SW exposure, plasma-Na⁺ levels rose significantly ($P < 0.05$) within all groups tested. However, no significant differences were detected between groups (Appendix Fig. 1). Mortalities due to SW exposure were low, ranging from 7 to 15 percent.

Gill Na⁺-K⁺ ATPase activities were similar for both groups from SSP but were significantly higher than those from the same brood of fish held in the hatchery ($P < 0.05$; Appendix Fig. 1).

Differences in ATPase activities between groups were not manifest in performance in the SW challenge tests. However, small sample sizes

Appendix 5 (continued)

not have yielded enough statistical sensitivity to detect significant differences.

Rock Creek

Blood-plasma Na⁺ concentrations of SW-exposed fish were significantly higher for the hatchery group than early and late groups reared at RC (Appendix Fig 1). Additionally, only fish from the hatchery exhibited a significant rise in Na⁺ level after the 24-h SW exposure ($P < 0.05$).

Mortalities were high at RC in one of the two replicates from Group 1 (67.9%). This was due to contamination of one of the containers used in transportation rather than from exposure of the fish to SW. Mortality of the other group in the replicate was zero. Mortalities in all other groups were low (3.2-5.0%).

ATPase activities at RC were not significantly different between off-station groups ($P < 0.05$; Appendix Fig. 1). However, observed ATPase levels at RC were higher than those of fish reared at SCNFH. ATPase activities did not rise above 20 micromoles Pi/mg prot/hr in any of the groups tested.

Summary and Conclusions

Results from both the SW challenge and the ATPase tests suggest that fish reared off-station, especially at RC, were in a more advanced stage of smoltification than those reared at SCNFH.

Appendix 5 (continued)

Future studies should monitor plasma-Na⁺ concentrations during 24-hr time periods at the beginning, midpoint, and end of off-station rearing periods to follow changes in plasma-Na⁺ regulating ability more thoroughly. Plasma-Na⁺ regulating ability would then be followed as progressive smolt development was occurring.

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social Security Pond

N-K ANALYSES

PLASMA Na⁺			
Between Groups (Fresh Na ⁺)	Hatchery (155.0)	Group 2 (155.0)	Group 1 (151.0)
Between Groups (Salt Na ⁺)	Hatchery (155.3)	Group 1 (155.0)	Group 2 (157.3)
Within Groups (Fresh-F, Salt-S)	Hatchery F S	Group 1 F S	Group 2 F S
ATPase ACTIVITY			
Group (ATPase)	Group 2 (29.5)	Group 1 (24.6)	Hatchery (13.0)
FORK LENGTH			
Group (FL)	Group 1 (78)	Group 2 (70)	Hatchery (68)

Rock Creek

N-K ANALYSES

PLASMA Na⁺			
Between Groups (Fresh Na ⁺)	Group 1 (155.1)	Hatchery (148.2)	Group 2 (157.1)
Between Groups (Salt Na ⁺)	Hatchery (155.7)	Group 1 (157.0)	Group 2 (144.7)
Within Groups (Fresh-F, Salt-S)	Hatchery F S	Group 1 F S	Group 2 F S
ATPase ACTIVITY			
Group (ATPase)	Group 2 (17.1)	Group 1 (16.4)	Hatchery (13.0)
FORK LENGTH			
Group (FL)	Group 1 (78)	Hatchery (69)	Group 2 (63)

Appendix Figure 1. Summary of results of a 24-h seawater challenge using release groups (group 1 - "early" fish; group 2 - "late" fish) of fed fish at Social Security Pond and Rock Creek and hatchery control fish from Spring Creek National Fish Hatchery. Bar graph represents blood-plasma Na⁺ levels in freshwater (F) at beginning of the challenge (open bar) and after a 24-h seawater (S) exposure period (hatched bar). ATPase values for each group are included at the base of the graph.

Summary tables represent the results of a Newman-Keuls multiple comparison including analyses of plasma -Na⁺ levels between groups and within groups, comparisons of ATPase activity among groups, and comparisons of sizes (fork lengths) of test groups.

Appendix 6. Summary of one-way ANOVA and planned comparison of size at release (fork lengths) of hatchery fish and early and late groups of fed fish reared in pens at Social Security Pond, 1985.

Early fish (Pens 7-12):

ANOVA:

<u>Source</u>	<u>D.F.</u>	<u>S.S.</u>	<u>M.S.</u>
Model	6	1848.2	308.0
Error	337	8363.6	24.8
Corrected total	343	10211.8	

Planned Comparison (Sokal and Rohlf 1969):

	<u>Pen Number</u>						<u>Hatchery</u>
	7	8	9	10	11	12	
EY =	3491	3570	3652	3412	3582	3560	2949
N =	50	50	50	50	50	50	45
Y =	69.8	71.4	73.0	68.2	71.6	71.2	65.5

$$SS = \frac{(2949)^2}{45} + \frac{(21,266)^2}{300} - \frac{(24,216)^2}{345} = 1122.11$$

$$F = \frac{S.S.}{M.S. \text{ Error (from ANOVA)}} = \frac{1122.1}{24.8} = \underline{\underline{45.21}}$$

$$F .05 (1,337) = 5.07$$

- Hatchery (control) significantly smaller than test groups.

Late fish (Pens 1-6):

ANOVA:

<u>Source</u>	<u>D.F.</u>	<u>S.S.</u>	<u>M.S.</u>
Model	6	351.2	58.5
Error	340	9935.1	29.2
Corrected total	346	10286.2	

Planned Comparison (Sokal and Rohlf 1969):

	<u>Pen Number</u>						<u>Hatchery</u>
	1	2	3	4	5	6	
EY =	3340	3385	3308	3350	3492	3334	2949
<u>N =</u>	50	50	50	50	50	50	45
Y =	66.1	67.7	66.2	67.0	69.8	66.7	65.5

$$SS = \frac{(2949)^2}{45} + \frac{(20,173)^2}{300} - \frac{(23,122)^2}{345} = 114.6$$

$$F = \frac{S.S.}{M.S. \text{ Error (from ANOVA)}} = \frac{114.6}{29.2} = \underline{\underline{3.92}}$$

$$F .05 (1,340) = 5.07$$

- No difference ($P > .05$) between hatchery (control) and test groups.

Appendix 7. Summary of one-way ANOVA and planned comparison of size at release (fork lengths) of hatchery fish and early and late groups of fed fish reared in pens at Rock Creek, 1985.

Early fish (Pens 1-6):

ANOVA:

<u>Source</u>	<u>D.F.</u>	<u>S.S.</u>	<u>M.S.</u>
Model	6	951.4	158.6
Error	334	12729.6	38.1
Corrected total	340	13860.9	

Planned Comparison (Sokal and Rohlf 1969):

	<u>Pen Number</u>						<u>Hatchery</u>
	1	2	3	4	5	6	
EY =	3438	3552	3441	3572	3703	3674	3116
N=	49	50	49	50	50	50	45
Y=	70.2	71.0	70.2	71.4	74.1	73.5	69.2

$$SS = \frac{(3,116)^2}{45} + \frac{(21,380)^2}{298} - \frac{(24,495)^2}{343} = 244.19$$

$$F = \frac{S.S.}{M.S. \text{ Error (from ANOVA)}} = \frac{244.19}{38.1} = \underline{\underline{6.41}}$$

$$F .05 (1,334) = 5.07$$

- Hatchery (control) significantly smaller than test groups 1

Late fish (Pens 7-12):

ANOVA:

<u>Source</u>	<u>D.F.</u>	<u>S.S.</u>	<u>M.S.</u>
Model	6	4584.5	764.1
Error	338	10705.4	31.7
Corrected total	344	15289.8	

Planned Comparison (Sokal and Rohlf 1969):

	<u>Pen Number</u>						<u>Hatchery</u>
	7	8	9	10	11	12	
EY =	3217	2862	3086	3365	3281	3117	3116
N=	50	50	50	50	50	50	45
Y =	64.3	57.2	61.7	67.3	65.6	62.3	69.2

$$SS = \frac{(3116)^2}{45} + \frac{(18,928)^2}{300} - \frac{(22044)^2}{345} = 1481.35$$

$$F = \frac{S.S.}{M.S. \text{ Error (from ANOVA)}} = \frac{1481.35}{31.7} = \underline{\underline{46.76}}$$

$$F .05 (1,338) = 5.07$$

- Hatchery (control) significantly larger than test groups.

Appendix 8. Summary of one-way ANOVA comparing size of early and late groups of fed fish in pens at release (fork lengths) at Social Security Pond and Rock Creek, 1985.

Social Security Pond

ANOVA:

<u>Source</u>	<u>D.F.</u>	<u>S.S.</u>	<u>M.S.</u>	<u>F</u>
Model	1	2207.9	2207.9	75.2
Error	597	17528.1	29.4	
Corrected total	598	19736.0		

$$F .05 (1,597) = 3.84$$

Rock Creek

ANOVA:

<u>Source</u>	<u>D.F.</u>	<u>S.S.</u>	<u>M.S.</u>	<u>F</u>
Model	1	11086.7	11086.7	261.3
Error	594	25200.3	42.4	
Corrected total	595	36287.1		

$$F .05 (1,594) = 3.84$$

Appendix 9. Weekly fish mortality by pen at Social Security Pond and Rock Creek, 1985.

		Date						
		4/18	4/25	5/2	5/9	5/16	5/23	5/30 6/5
<u>Social Security</u>								
(fed fish)								
Pen Number								
1				3	91	0	18	(5/16 - release
2				0	53	1	17	Pens no. 7-12)
3				3	15	0	8	
4				0	37	0	25	(5/17 - release
5				0	211	0	2	Pens no. 1-6)
6				1	103	1	2	
7	632	4	2	0	2			
8	189	4	0	1	4			
9	133	1	1	7	5			
10	44	4	0	0	2			
11	53	3	0	0	5			
12	103	10	0	15	8			
<u>Rock Creek</u>								
(fed fish)								
Pen Number								
1	66	0	1	0	1	2		
2	92	1	24	1	0	9		
3	192	1	13	0	3	5		(5/20 - release
4	121	1	9	0	3	4		Pens no. 1-12)
5	190	1	11	1	6	10		
6	382	0	11	0	15	14		
7		1	0	35	2	17		
8		0	0	65	0	5		
9		0	2	49	1	19		
10		4	0	39	0	17		
11		0	2	30	1	6		
12		0	1	35	5	27		
(unfed fish)								
13 - low density			0	0	0	6	7	7
14 - low density			0	0	1	9	5	33
15 - medium density			0	0	1	16	37	33
16 - medium density			0	0	0	18	161	134
17 - high density			0	0	13	80	758	257
18 - high density			0	0	30	51	507	233

(6/5 - release
Pens no. 13-18
into barrier
net)

Appendix 10. Results of comparison of sizes (fork lengths - mm) and ATPase activities of unfed fish at low (.002 lb/ft³), medium (.004 lb/ft³), and high (.008 lb/ft³) densities using a one-way analysis of variance and a Newman-Keuls test of multiple comparison, 1985.

Summary of one-way ANOVA (P = .05):

<u>Dependent variable</u>	<u>Source</u>	<u>d.f.</u>	<u>ANOVA S.S.</u>	<u>f-value</u>	<u>P > f</u>
Fork length	Density	2	693.44	17.09	0.0001
ATPase activity	Density	2	170.73	4.43	0.0164

Summary of multiple comparison^a

<u>Variable</u>	<u>Density</u>	<u>Mean Value</u>	<u>N</u>	<u>Grouping</u>
Fork length	Low	63.9	98	A
	Medium	62.9	100	A
	High	60.3	100	B
ATPase activity	Low	14.97	19	A
	Medium	13.39	20	AB
	High	10.84	20	B

^aThose with same grouping number were not significantly different at .05 level.

Appendix 11. Size distribution and taxonomic composition of fish predators collected within the barrier net during two seine hauls on 5/9 and 5/22/85, Rock Creek (total lengths).

<u>Species</u>	<u>Length frequency (mm)</u>					<u>Total</u>
	<u>150-200</u>	<u>200-250</u>	<u>250-300</u>	<u>300-350</u>	<u>350-400</u>	
Largemouth bass	1	0	2	1	0	4
Smallmouth bass	7	3	2	0	1	13
Crappie	85	38	0	1	0	123
Yellow perch	10	1	0	0	0	11
Squawfish	1	0	0	1	0	2

Appendix 12. Size (length, weight, and number/lb) and gill ATPase activity of control groups of fall chinook salmon reared at the Little White Salmon and Spring Creek National Fish Hatcheries, 1985.

<u>Little White Salmon</u>	<u>Date</u>				
	<u>5/7</u>	<u>5/17</u>	<u>5/31</u>	<u>6/12</u>	<u>6/19</u>
Length (mm)	62.0	63.7	69.0	73.2	75.9
Weight (g)	2.43	2.84	3.73	4.32	5.16
No./lb	187	160	122	105	88
ATPase activity	9.53	-		8.93	7.28

<u>Spring Creek</u>	<u>Date</u>							
	<u>4/8</u>	<u>4/17</u>	<u>4/24</u>	<u>5/2</u>	<u>5/8</u>	<u>5/15</u>	<u>5/31</u>	<u>6/12</u>
Length (mm)	45.0	46.5	48.8	57.7	61.6	65.8	71.7	76.3
Weight (g)	1.20	1.33	1.49	2.20	2.57	2.67	3.66	4.44
No./lb	378	341	305	225	177	170	124	102
ATPase activity	9.0	11.3	7.4	11.2	11.0	9.1	16.0	9.5

Appendix 13. Efficiency ratios (E.R.) Senn et al. 1984) for off-station rearing using net pens, a barrier net, and traditional hatchery methods using concrete raceways. Off-station costs were determined from actual expenses incurred using various test densities (Novotny et al. 1985) and from estimated expenses using recommended loading densities for net-pen rearing (Senn et al. 1984). Hatchery costs were determined from a combination of summarized expenses (Senn et al. 1984) and actual costs of rearing fish at the Little White Salmon National Fish Hatchery (J. Bodle, Hatchery Manager). Discounting rate (r) was considered to be .10 for these calculations, but it may vary considerably.

A. Net pen-low density (.043 lb/ft³):

$$E.R. = \frac{1000A_{20,r}}{20241 + \text{initial cost (pens, frames, feeders)} + 4791 + \text{initial cost (nets, covers)} + 4791V_{7,r} + \text{replacement cost} + 4791V_{7,r} - \text{replacement cost} + 684V_{7,r} + \text{unused net \& cover} + 94A_{20,r} + \text{food cost} + 1252A_{20,r} + \text{O \& M} + 2417A_{20,r} + \text{labor cost}} = \frac{1b/\$}{.14} \quad \frac{\$/1b}{7.14}$$

B. Net pen-high density (.086 lb/ft³):

$$E.R. = \frac{1000A_{20,r}}{10203 + \text{initial cost (pens, frames, feeders)} + 2415 + \text{initial cost (nets, covers)} + 2415V_{7,r} + \text{replacement cost} + 2415V_{7,r} - \text{replacement cost} + 345V_{7,r} + \text{unused net \& cover} + 30A_{20,r} + \text{food cost} + 631A_{20,r} + \text{O \& M} + 1218A_{20,r} + \text{labor cost}} = .26 \quad 3.85$$

C. Net pen (.3 lb/ft³) Senn et al. 1984):

$$E.R. = \frac{1000A_{20,r}}{2915 + \text{initial cost (pens, frames, feeders)} + 690 + \text{initial cost (nets, covers)} + 690V_{7,r} - \text{replacement cost} + 690V_{7,r} + \text{replacement cost} + 98V_{7,r} + \text{unused net \& cover} + 300A_{20,r} + \text{food cost} + 180A_{20,r} + \text{O \& M} + 348A_{20,r} + \text{labor cost}} = .73 \quad 1.37$$

D. Barrier net (.001 lb/ft³):

$$E.R. = \frac{1000A_{10,r}}{.25,200 + \text{initial cost (netting)} + 10,714 + \text{initial cost (anchors, floats, etc.)} + 1796A_{10,r} + \text{O \& M}} = .13 \quad 7.69$$

E. Hatchery comparison (single pass):

$$E.R. = \frac{1000A_{50,r}}{6440 + \text{raceway cost} + 6000 + \text{plumbing cost} + 6000V_{25,r} + \text{replacement cost} + 631A_{50,r} + \text{water cost} + 600A_{50,r} + \text{food cost} + 2350A_{50,r} + \text{O \& M} + 835A_{50,r} + \text{labor cost}} = .18 \quad 5.56$$

Summary of Expenditures for 1985

1.	Salaries and Benefits	145,611
2.	Travel and Transportation	15,611
3.	Equipment	
	Non-expendable	47,919
	Expendable	19,110
4.	Operation and Maintenance	8,389
5.	Overhead	111,896
		<hr/>
	Total	348,536