

48<sup>th</sup> Annual  
Pacific Northwest  
Fish Culture Conference



SH  
151  
.N67  
1997

*Proceedings*

December 2-4, 1997

Salishan Lodge  
Gleneden Beach, Oregon



PROCEEDINGS  
OF THE  
FORTY-EIGHTH  
NORTHWEST FISH  
CULTURE  
CONFERENCE

DECEMBER 2 - 4, 1997

SALISHAN LODGE  
GLENEDEN BEACH, OREGON

CHAIR  
R.Z. SMITH  
NATIONAL MARINE FISHERIES SERVICE

## The Northwest Fish Culture Conference

The Northwest Fish Culture Conferences are annual, informal meetings held for the free exchange of current information and ideas concerning all areas of fish culture. These conferences are hosted on a rotating basis by the various fisheries agencies and entities of the Northwest. At the conferences, progress reports of management practices and problems, new development, and research studies are presented. Both within the meeting and outside the meeting setting, active discussion, constructive criticism, and personal contacts are not only encouraged but actively cultivated. All persons interested in or associated with fish husbandry are invited to attend and to actively participate. Students from the Northwest's colleges and junior colleges are encouraged to attend and interact with the professionals in the field of fish culture. The subject matter of the conferences is limited to topics that have direct application to fish culture.

These "Proceedings" contain unedited, non-peer reviewed briefs of oral reports presented at the conferences. Although care was taken to present the information in an accurate manner, the "Proceedings" were prepared in-house by fisheries biologists not by a professional publishing house. Errors, both in interpretation and typing, may have slipped passed review and, in advance, we apologize for any confusion that might arise. Additionally, much of the material presented involves progress of incomplete studies or reports. **THESE INFORMAL RECORDS SHOULD NOT BE INTERPRETED OR QUOTED WITHOUT FIRST CONTACTING THE PRESENTERS.**

Mention in these "Proceedings" does not indicate approval, recommendation, or endorsement by the National Marine Fisheries Service or the other participating fisheries entities of any proprietary product or proprietary material.



# *48<sup>th</sup> Annual Pacific Northwest Fish Culture Conference*

PREFACE

Approximately 400 enthusiastic participants with diverse backgrounds but with a common interest in fish husbandry attended the Forty-Eighth Annual Northwest Fish Culture Conference, in Gleneden Beach, Oregon. In this time of budget cut-backs being experienced by most fishery agencies, I feel that this represents a significant commitment of time and money and shows the importance that is placed on the value of this unique conference. Once again the meeting facilities and meals provided by Salashan Lodge were unparalleled. These accommodations added to the experience of those attending the conference while they spent three days being exposed to "state-of-the-art" presentations on topics ranging from new natural rearing strategies to fish culture in Russia. Although these presentations are the backbone of the conference, a benefit that is at least as important as the valuable information received in the meeting room is the information exchanged and the contacts made during breaks, in the halls, and during after-hours "events."

One thing that really made the conference special and tied everything together through the theme of "Practical Fish Culture" was the outstanding art print that was designed for the conference by Kathryn Kostow. Kathryn, Genetics Program Leader for Oregon Department of Fish and Wildlife's Natural Production Program, used her artistic skills as well as her knowledge of Northwest art to develop "The People Producing Salmon." As detailed in her short description of the background for this art work (included on page iv of these Proceedings), it represents a team effort between the human people and the salmon people to produce salmon. I would like to thank her again for all her work and "creative genius" which added to the overall conference experience.

Commercial exhibitors (listed on pages 191-192 of these Proceedings) provided displays of the latest in fish culture equipment and technology. They graciously hosted the coffee breaks. This was greatly appreciated and significantly added to the opportunities for personal contacts during the conference. They also provided prizes for the door prize drawings held throughout the conference. A list of prizes, donors, and winners are listed on pages 193-196. I would like to give special thanks to the feed companies for providing snacks during our hospitality hour.

Planning and implementing a conference such as this one is a major undertaking. Without the support of the personnel from the National Marine Fisheries Service's Hatcheries and Inland Fisheries Branch in Portland, Oregon, I never could have done it. I would like to thank Steve Smith, Branch Chief, for making staff time available in the face of an overwhelming Endangered Species Act work load. I would especially like to thank Mike Delarm, Robert Bayley, Lance Kruzic, Herb Pollard, and Derrick Poon for all their work in preparation for the conference as well during registration. I would also like to thank them for serving as Session Chairs during the conference. Special thanks go to Robert Bayley for his work on the Program and Lance Kruzic for his work with the audio-visual equipment. I would like to thank Dr. Rich Holt, Trent Stickell, and George Nandor from the Oregon Department of Fish and Wildlife for serving as Session Chairs. Tom Rodgers and Margaret Whipple from the Idaho Department of Fish and Game (the hosts for next year) provided valuable assistance with registration. Last, but certainly not least, I would like to give special thanks to Darlene Pollard, Herb's better half, for volunteering to help with registration and anything else necessary to make the conference run smoothly.

A final thanks needs to be given to those who made presentations. For most of us, it is not an easy task to get up in front of a large group to give a presentation. It represents a significant commitment of time and effort and all presentations were well received.

My only disappointment deals with these Proceedings. Unfortunately, a number of speakers chose not to prepare papers to be included in the Proceedings. This will reduce the value of the Proceeding to document the information provided. I hope this does not become a trend.

I have enjoyed working on this project and I am looking forward to the 1999 meeting in Boise, Idaho on December 1-3. This is the third conference that I have been involved with hosting and, although I wouldn't have missed it for the world, the next time it is the National Marine Fisheries Service's turn to host it, I will be retired and will attend as a civilian!!

R.Z. Smith



# *"The People Producing Salmon"*

By Kathryn Kostow

*"The People Producing Salmon" is inspired by the art of Northwest Native Americans. It represents a team effort between the human people and the salmon people to produce salmon. Since it was created for the 1997 Northwest Fish Culture Conference, it can be taken as a literal "production" of salmon. Alternatively, it can represent the broader effort underway in the Northwest to improve conditions for, and therefore the production of, salmon.*

*The salmon people are represented by a ripe father salmon (in blue) and mother salmon (with red eggs). They carry in their bodies the essence of their peoples spirits, represented by the faces in their eyes and hearts. Of course, the salmon people are the star players in the production of salmon.*

*The human hands, passing eggs and milt, represent the human involvement in the production of salmon. But the human face in the center looks both concerned and uncertain. In the head of the human is the concept of a salmon, but the concept is confused and incomplete. These aspects represent the concern, but also the debate and uncertainty, inherent in our efforts to improve Northwest salmon populations.*

# Table of Contents

Preface .....	iii
"The People Producing Salmon" .....	iv
Keynote Address: Managing Salmonid Hatchery Programs into the Future .....	vii
Session I— Nutrition .....	1
Are High Fat Diets Affecting the Appetites of Juvenile Salmonids? .....	3
Do We Need to Feed Hatchery Fish in the Winter? The Effect of Low Temperature and Fasting on the Growth, Metabolism, and Smoltification of Coho Salmon .....	5
A method for Assessment of the Efficacy of Feeding Attractants for Fish .....	7
Development of Open Formula Diets and New Feeding Strategies: A Progress Report .....	15
Evaluation of the Influence of Diet and Demand Feeding on Fish Performance and Phosphorus Discharge .....	29
Session II— External Influences .....	31
A Trout's Perspective: Migration Patterns of Volitionally Release Hatchery Trout in the Elochoman River .....	33
Pinniped Scarring at Beaver Creek, a Lower Columbia River Hatchery .....	39
Movement of Radio-Tagged Cowlitz River Hatchery Winter Steelhead .....	45
Stream Nutrient Enhancement with Hatchery Diverted Salmonid Carcasses .....	47
The Suquamish Tribe's Approach to Successful Chum Salmon Enhancement .....	49
Session III— New Natural Rearing Strategies .....	57
Predator Avoidance Training Can Increase Post-release Survival of Chinook Salmon .....	59
Chemical Alarm Signaling in Chinook Salmon Smolts: An Opportunity for Anti-predator Conditioning .....	63
Addition of Floating and Bottom Structures to Concrete Raceways at Solduc Hatchery .....	69
Evaluation of Semi-Natural Rearing for Coho Salmon .....	71
Evaluating the Effects of Complex Rearing Habitats on Juvenile Fall Chinook .....	79
Seminatural Raceway Habitat Increases Chinook Salmon Post-release Survival .....	81
Session IV— Pathology/Disease .....	93
Results from a Chloramine-T Clinical Efficacy Trial to Control Mortality Among Fall Chum Salmon Caused by Bacterial Gill Disease .....	95
The Use of Penicillin-G for Control of Bacterial Coldwater Disease in Salmonid Fishes .....	101
Culling of Eggs from BKD Positive Spring Chinook in Oregon .....	103

Integrated Management of Bacterial Kidney Disease in the IDFG Hatchery Program .....	109
Session V— Captive Broodstock .....	113
Cryo-preservation of Salmonid Sperm .....	115
Synchronized Spawning of Wild and Captive Broodstock .....	117
An Overview of the Captive Broodstock Program in NE Oregon .....	123
Redfish Lake Sockeye Salmon Captive Broodstock Program, NMFS .....	127
Dungeness Chinook Freshwater Captive Broodstock Program .....	137
White River Spring Chinook Rebuilding Program .....	139
Session VI— Hatchery Practices .....	143
State-of-the-Art Aquaculture Techniques .....	145
“Taming the Beast”: The Road to Fast, Easy, Simple Computerized Feed Programming .....	147
Use of Electronic Tag Detectors to Separate Mass-Marked (Adipose-Clipped Only) Coho from CWT Coho at WDFW Facilities in 1997 .....	151
Environmental Compliance Audits at Fish Hatcheries .....	155
Helpful Hints and Tips of the Trade .....	159
Investigation of Rearing and Release Strategies Affecting Adult Production of Spring Chinook Salmon .....	161
Practical Fish Culture at Grovers Creek Fall Chinook Salmon Hatchery .....	173
Aquaculture in Far East Russia, Kamchatka Peninsula, and Sakhalin Island .....	185
Posters .....	187
Propagating Juvenile Fall Chinook Salmon in Michigan Raceways at Umatilla Hatchery .....	189
Commercial Exhibitors .....	191
Door Prize List .....	193
Northwest Fish Culture Conference Historical Record .....	197

# Key Note Address: Managing Salmonid Hatchery Programs into the Future: A National Marine Fisheries Service Perspective

William L. Robinson

*National Marine Fisheries Service, Assistant Regional Administrator for Sustainable Fisheries,  
7600 Sand Point Way NE, Seattle, WA 98115-0070*

## Introduction

For those of us who call ourselves fisheries managers, and I include all of those who work in the area of salmonid hatcheries and artificial production as fisheries managers, these are challenging times to say the least.

West Coast salmon stocks are at their lowest point in history; many are either already extinct or are rapidly reaching that point.

The reasons behind this precipitous decline are numerous and include the usual suspects: Logging, mining, grazing, irrigation, road construction, urban development, and hydropower activities have combined to result in what can only be described as a long-term, relentless assault on the quantity and quality of freshwater habitat vital to the production of indigenous, naturally spawning salmonids.

Hatchery production and over-harvest also carry their fair share of the blame. In particular, the early success of hatchery programs in the 60's and 70's, riding on the crest of productive ocean conditions, almost certainly created unreasonable expectations for the future, and, in hindsight, were conducted without an adequate science-based understanding of the risks to wild populations.

Although not deliberate, too little thought by fisheries managers went into the consequences to wild salmon stocks of harvest strategies in mixed-stock fisheries that were intended to fully harvest surplus hatchery returns. It is now well known and accepted that wild stocks must be harvested at significantly lower harvest rates than most hatchery stocks can withstand in order to fully seed available spawning habitat.

Even less well understood was the potential loss of genetic diversity, loss of survival fitness, and other ecological effects resulting from straying and interbreeding of both non-indigenous and indigenous hatchery stocks with indigenous wild stocks.

Those of you listening carefully may have noticed that I have not spread the blame to severely reduced ocean productivity and survival that many of our salmon stocks have had to endure for the last two decades. There is no doubt that poor ocean survival has drastically impacted all salmon stocks, both hatchery and wild. But, frankly, I am reluctant to focus on ocean productivity as a primary cause for salmon decline because it is too convenient as a reason to not face up to the myriad of human-caused factors for the salmon's decline.

The evolution of wild salmon has resulted in life histories that include run-timing and other characteristics that bring great resiliency to environmental conditions. Wild salmon have utilized that resiliency to persist and recover despite natural disasters and cyclic unfavorable ocean conditions. To ignore or even to delay directly attacking all of the human-caused factors for salmon decline and hope that a change in ocean productivity will make everything all right is a sure recipe for continued extinctions.

The hatchery role is evolving to include a conservation role for native fish. In addition, we now have a better understanding of the importance of wild fish. This means that the hatchery managers will need to adjust to these new and changing management priorities. Much of my talk will focus on the change in management priorities and to identify some of the issues behind the drive for hatchery change and reform.

## NMFS Role and How NMFS, Northwest Region has Reorganized to Fulfill that Role

Under a Memorandum of Agreement with the U.S. Fish and Wildlife Service, NMFS has the responsibility for administering the Endangered Species Act for all anadromous salmonid species. NMFS, along with the Pacific Fisheries Management Council, is responsible for managing the West Coast ocean salmon fisheries for 3-200 miles. In addition, we administer and fund numerous grant programs for data collection and research, as well as plan and conduct a substantial research program by our Northwest Fisheries Science

Center. NMFS also administers the Mitchell Act Hatchery Program in the Columbia River.

NMFS Northwest Region has reorganized recently to better fulfill its responsibilities. We have created four distinct programs, each reporting directly to the Regional Administrator, Will Stelle.

1. **Hydropower Program**—Fulfills our ESA and other responsibilities such as FERC relicensing for hydropower development, and in particular monitors and participates in the implementation of the multi-year Biological Opinion on the Columbia River Hydropower System.
2. **Protected Resources Division**—Administers the listing and recovery activities under the ESA.
3. **Habitat Division**—Administers all ESA Activities related to habitat including Section 7 consultations for Federal activities and Section 10 activities for non-Federal lands.
4. **Sustainable Fisheries Division**—The only Northwest Region Division with multiple missions and responsibilities. Under the ESA, NMFS has grouped the factors for the decline of salmon under what we call the 4H's; hydropower, habitat, harvest, and hatcheries. The Sustainable Fisheries Division administers all ESA activities, including Section 7 consultations and Section 10 permit activities for two of the 4H's; harvest and hatcheries. In addition to our ESA mission, the Sustainable Fisheries Division also must exercise the Governments' Federal treaty Indian trust responsibilities are defined in the Treaties of 1855 and subsequent court cases as well as in other treaties. Finally, we believe the ultimate objective for salmon recovery is to have viable commercial and recreational fisheries for all.

#### **What is NMFS' Current Perspective on Hatchery Production?**

Although we admit that we don't have all the answers yet, we basically believe it is time to reexamine all of our hatchery programs with the objective of first reducing or preventing adverse impacts on indigenous, wild salmonids, then utilizing existing capacity, to the extent

necessary, to support the recovery of depressed wild stocks, and finally to become more cost-effective and performance, or results, oriented.

#### **Mitigation and Harvest Augmentation**

Fisheries managers now have a better understanding of the importance of wild fish and the need to design programs and manage our hatchery programs consistent with maintaining healthy wild populations. The purpose of the lion's share of traditional hatchery production in the Pacific Northwest has been to mitigate for lost habitat and production and to provide additional harvest opportunity. Based on early successes with a variety of stocks, many programs relied on a single stock, often non-native to the area, and spread that stock over wide geographical areas. The major concern with the practice, obviously is high levels of straying and interbreeding with native wild stocks resulting in loss of genetic diversity, loss of survival fitness (adaptability) of the native populations, and other ecological effects.

To reduce these adverse impacts, hatchery program managers may need to take drastic steps such as reducing production levels, changing brood stocks from non-native stocks to stocks native to the geographic area, or isolating hatchery populations of non-native stocks in order to reduce straying and interbreeding.

Hatchery populations based on native stocks carry the same potential risks to wild spawning populations if the brood stocks have been isolated from the wild population for too long a time. Consideration might be given to regularly infusing the hatchery brood stock with wild spawners in order to prevent genetic divergence and reducing the effects of straying.

In the future, hatchery managers may need to revise the paradigm for measuring success. Overall percent return or contribution to fisheries may be less important than maintaining programs that continue to provide harvest opportunity and brood stock, but are simultaneously "wild stock friendly." It is possible that the number of hatchery fish released can be reduced by increasing survival through rearing techniques that more closely resemble the natural environment, especially in years of low productivity.

A final thought on the subject of mitigation. It will continue to play an important management role.

## Managing Salmonid Hatchery Programs into the Future

However, in some cases, mitigation-based hatchery production will have to be reduced in order to prevent adverse interactions with native stocks. In those cases, those mitigation dollars should be redirected to address habitat, passage, or other factors for decline.

### Wild Stock Preservation and Recovery

Hatchery programs can contribute to the restoration and recovery of native, wild stocks and I expect that there will be a growing need to utilize both existing and new hatchery capacity for this purpose.

Because of the genetic and ecological risks of hatchery programs to wild stocks, NMFS Interim Policy on Artificial Production strongly endorses utilizing hatcheries for wild stock recovery only where there is no other alternative, and then only with indigenous, native stocks unless there is absolutely no other choice.

In the extreme, captive brood programs may be required to prevent the extinction of remnant populations. Supplementation programs with either adults or juveniles may be established to give wild populations a "jump start" towards recovery. These types of programs are generally still unproven and in the experimental stages, although they may eventually play an important role in recovery. However, it should be obviously clear, that these types of programs are doomed to failure unless the primary factors for decline are addressed and fixed. Neither should be considered a long-term substitute for healthy native populations.

Nevertheless, artificial production does offer potential benefits to wild stock recovery. It can: (1) Reduce short term extinction risk; (2) Maintain populations while factors for decline are addressed; (3) Establish reserve populations for future augmentation if necessary; (4) Be used to reseed vacant habitat; and (5) Provide scientific information to better understand the role of hatcheries and wild stock production. Before any new programs are initiated, however, we believe a full risk/benefit analysis should be conducted. In the final analysis there is no substitute for healthy natural populations.

### Cost Effectiveness and Performance Based Decision Making

The final area of concern is how to make our hatchery programs maximally effective in an era of uncertain funding. We believe state, tribal, and Federal hatchery program managers should work together to develop decision-making criteria to enable them to make funding decisions based on the performance results and cost effectiveness of each individual program.

### Summary

- Hatchery Programs will continue to play a significant role in the Pacific Northwest.
- Hatchery programs can represent a significant risk to native populations. This risk must be considered and managed to promote healthy native populations.
- The hatchery role is evolving to include native stock restoration in addition to the traditional mitigation/fishery enhancement role.
- The role of hatcheries will vary depending on quality, quantity, and the future prospects for recovery of fish habitat in a watershed.
- Adequate monitoring and evaluation is critical so that managers can have a better understanding of hatchery and wild fish interactions.
- Hatchery programs cannot be considered in isolation. Consideration must also be given to harvest regimes and habitat.
- Hatchery programs must be evaluated on their performance in relation to the changing needs within the region.

Robinson

# Session I

## **Nutrition**

Session Chair:

Dr. Derek Poon  
(National Marine Fisheries Service)



# Are High Fat Diets Affecting the Appetite of Juvenile Salmonids?

Dr. Karl Shearer <sup>1</sup>

*National Marine Fisheries Service*

Jeffrey Silverstein

*University of Washington, School of Fisheries*

**Abstract—** Studies designed to examine the role of adiposity (body fat level) on regulation of feed intake in fish have generally used starvation to produce fish with different adiposities. This has led to a confounding of the role of adiposity and starvation-induced compensatory growth on feed intake. Two experiments were conducted to determine if adiposity affected feed intake in juvenile chinook salmon (*Oncorhynchus tshawytscha*) with different nutritional histories. Fry were fed high fat (23%) or low fat (3%) diets at high (satiation) and low (one-half satiation) ration levels for seven months prior to the start of the experiment. This pre-treatment produced fish averaging 22 g with 11.3% (high fat diet) and 5.4% (low fat diet) body fat when fed to satiation or 11 g with 8.1% (high fat diet) and 4.0% (low fat diet) body fat when fed at one-half satiation. Experiment I was a 2 x 2 factorial design where duplicate groups of 20 fish from the 22 g groups were fed high (20.3%) or low (2.5%) fat diets twice daily to satiation six days/week for 3 weeks. Daily feed intake was recorded. The same protocol was used in Experiment II using fish (40 fish/tank) from the slower growing 11g groups. Feed intakes on day one and cumulative feed intakes after 21 days were compared using Two-Way ANOVA with initial whole body fat and dietary fat as the independent variables. Effects were considered significant at  $P < 0.05$ . In both experiments, high body fat and low dietary fat led to significantly lower feed intake on day one. After 21 days of feeding however, only the effect of high body fat was significant, indicating that fish adjusted to the low fat diets. Our results show, that in both fast and slow growing juvenile chinook salmon, that adiposity plays a role in regulation of feed intake as it does in other vertebrates.

---

<sup>1</sup> Karl elected to only have the abstract of his talk published in the Proceedings.

## Are High Fat Diets Affecting the Appetite of Juvenile Salmonids?

# **Do We Need To Feed Hatchery Fish In The Winter. The Effect Of Low Temperature And Fasting On Growth, Metabolism And Smoltification Of Coho Salmon**

Donald A. Larsen <sup>1</sup>, Brian R. Beckman, and Walton W Dickhoff  
*National Marine Fisheries Service*

**Abstract**— Recent research in our laboratory has provided evidence that both successful hatchery smolts and wild smolts display a more dramatic seasonal dynamic in growth, metabolism and endocrine physiology prior to smolting, compared with less successful hatchery fish. This dynamic is characterized by low growth in winter and high growth in the spring, prior to outmigration, and dramatic changes in various physiological parameters including condition factor, liver glycogen, body lipid+plasma thyroxine (T4), plasma insulin-like growth factor I (IGF-I), plasma insulin, and gill Na K ATPase. In the transition from autumn to winter, wild fish shift from an anabolic (energy storage) to a catabolic (energy mobilization) state characterized by decreased feeding, a reduction in metabolic reserves and reduced growth rate. In the early spring the pattern is reversed by a switch from catabolism to anabolism with feeding, energy stores, and growth rate all increasing prior to outmigration. We have hypothesized that this physiological dynamic is an essential process seldom experienced by hatchery fish which are commonly fed throughout the winter months.

The objective of this study was to examine the effect of winter feeding and fasting, under both high (10°C) and low (2.5°C) temperature, on growth and physiology of coho salmon prior to and during smoltification (Jan-May). The temperatures were intended to approximate typical winter hatchery ground water (10°C) and surface water (2.5°C) conditions. The treatments consisted of the following groups: Warm-Fed (WF), Warm-Not Fed (WNF), Cold-Fed (CF) or Cold-Not Fed (CNF). During the five months (Oct-Feb) prior to smoltification fish were either fed (at the manufacturers specified rate at each temperature 1.5% BW-10°C, 0.7% BW-2.5°C) or starved during a two month period (Jan-Feb). During March through May, all groups were reared at 10°C and fed at 1.5% BW. Throughout the investigation the following parameters were (or will be) measured: length, weight, instantaneous growth, hepatosomatic index, body lipid, liver glycogen, gill Na K ATPase and plasma levels of IGF-I, T4, and insulin.

The results show that WF fish grew continuously throughout the winter and were larger than the other treatments. All other groups were smaller and showed depressed growth during Jan and Feb; including the CF group, despite the fact that it was fed. Among the physiological parameters which have been measured to date; condition factor, hepatosomatic index, plasma IGF- I, plasma insulin and liver glycogen were all highest in the WF fish and depressed in the WNF fish during the winter. The CF and CNF groups were intermediate. However, during the spring, when all groups were fed and returned to the warm temperature, the previously starved groups (WNF and CNF) showed very similar, dynamic changes in instantaneous growth and most physiological parameters while the continuously fed groups (WF and CF) displayed less change.

The data are not yet complete, and we do not know whether winter-starved fish show smolt development equivalent to the winter-fed fish. The fish were not tested for migratory performance. However, the growth and physiological profiles of the winter-starved animals, at both high and low temperature, more closely resembled that of wild salmonids. Future efforts to rear more "wild like" salmonids under hatchery conditions should recognize the importance of the interaction between season, temperature and feeding on the physiology of the fish being released.

---

<sup>1</sup> Don elected to only have the abstract of his talk published in the Proceedings.

## Do We Need To Feed Hatchery Fish In The Winter?

# A Method for Assessment of the Efficacy of Feeding Attractants for Salmonid Fish and Their Effect on Feeding Behavior

C.K. Oikawa and B.E. March

Department of Animal Science, University of British Columbia, Vancouver, B.C.

**Abstract-** A method is described for evaluating the feeding response of juvenile rainbow trout, *Oncorhynchus mykiss*, to feeding attractants. The attractants were liquid hydrolysates of krill, *Euphausia pacifica*, and were either mixed into the diet before pelleting or coated on the surface of the formed pellets. Fish were fed once daily to satiation. Feed consumption and wastage were recorded in 1 minute intervals. The use of feeding stimulants increased the amount and rate of feed intake and decreased feed wastage. Coating the stimulant on the pellet surface enhanced the effects. Differences in feeding response were observed in the first minute of feeding and after five days of feeding. These differences were maintained throughout the experiment suggesting that the effects of feeding stimulants are long-term.

## Introduction

The use of flavor enhancers in fish feeds could provide several benefits to fish culturists. Improving the flavor of fish diets can increase feed intake and may be particularly helpful for specialized feeding situations such as encouraging juvenile fish to initiate feeding, and increasing the acceptability of medicated feed (Toften et al, 1995). Feeding stimulants may be especially important for fish diets of the future. Current fish diets use fishmeal as a major protein source and future supplies of this ingredient are not expected to meet projected demand. Therefore future diets will likely incorporate higher levels of plant protein products than are currently used. Plant material, however, is not normally eaten by carnivorous fish and can be unpalatable (Rumsey, 1986; Teskeredzic et al., 1995).

Fortunately, fish can be induced to bite and/or swallow normally unpalatable objects such as cotton balls if they are appropriately flavored (Jones, 1989). This is because taste and smell are important senses for feeding behavior and the recognition of food. Fish use their sense of smell to help locate food and the sense of taste is involved in the final decision to either reject or swallow an object in the mouth (Appelbaum et al., 1983).

Feeding stimulants are often associated with natural food sources. When researchers have analyzed certain palatable foods such as shrimp, they have found that the ingredients responsible for the attractive flavor are typically the free L-forms of certain amino acids, and nucleotides (e.g. Carr & Chaney, 1976). Feeding stimulants act in complex ways. When separate feeding stimulants are combined, the mixture can result in either the enhancement or cessation of feeding (Adron & Mackie, 1978), and therefore it is important to test a potential feeding attractant in the diet for which it is intended.

The following paper presents data from three feeding trials conducted at the University of British Columbia. Although the trials were run to test a method for the evaluation of feeding stimulants this paper will focus the general effects feeding stimulants may have on the feeding behavior of salmonid fish.

## Methods and Materials

Three diets were used. A control diet was formulated to contain a relatively high concentration of plant material (62%) as a protein source with the remainder of protein supplied by fish meal (20%). The control diet contained, on an as fed basis, fish meal (20%), ground whole wheat (20%), canola meal (22%), soybean concentrate (10%), corn gluten meal (10%) herring oil, stabilized with ethoxyquin (13%), sodium lignosulfonate (2%), and a vitamin & mineral premix (3%). Half of the fish oil was mixed into the diet prior to pelleting and the remainder was sprayed onto the pellets. The test ingredient (2%) was either a hydrolysate of krill, *Euphausia pacifica*, (experiments 1 & 2) or acid preserved krill (experiment 3 only). The krill was added in substitution for a dry weight equivalent of 2% of the fishmeal in the control diet. Stimulants were either mixed into the diet before pelleting (stimulant diet), or coated on the outside of the formed pellets (stimulant coated diet).

Rainbow trout (*Oncorhynchus mykiss*) were randomly distributed to nine treatment tanks (150 L) with individual flow through (2L/minute) of dechlorinated municipal freshwater. Each diet was randomly assigned to each of three replicate tanks. The culture conditions are shown Table 1. The fish were maintained on a 12 hour photoperiod.

Fish were fed, one tank at a time, once daily to satiation (4 minutes experiments 1 & 2; 4-11 minutes experiment 3). The order of feeding followed the random allotment

**Table 1. Culture Conditions for Experiments 1-3.**

Parameter	Exp. 1	Exp. 2	Exp. 3
Initial Fish Weight (g)	10.4	19.5	19.0
Fish/ 150 l tank	95	58	81
Water temperature (C°)	5.5-7.5	8.5-10.5	13.0-14.0
Feeding trial length (days)	30	30	19

of dietary treatments so that replicates of a given treatment were not fed successively. Water flow was turned off during feeding to eliminate differences between tank water flow patterns, and to prevent uneaten pellets from draining away. Feed was dispensed with a spoon to prevent skin contact and contamination of flavor. Diets were coded to conceal their identity.

Feed was dispensed as rapidly as the fish would eat, and feeding rates were adjusted as feeding slowed to minimize feed wastage. Feed consumption and wastage were determined for each successive minute by feeding from a separate container in each minute and counting the uneaten pellets at the tank bottom at the end of each minute. Wastage weight was estimated by multiplying pellets numbers by average pellet weight. The resulting weight was subtracted from the weight of feed dispensed to estimate feed consumption.

Data were analyzed with Analysis of Variance and Tukey's HSD tests, both at the 0.05 significance level using Systat software v. 5.02. Feed wastage data were transformed using a modified arcsine (Freeman-Tukey) transformation.

### Results

In all experiments, fish fed the diets containing the stimulant ate more than the fish fed the control diet (Figure 1). Coating the pellet with the stimulant enhanced these effects in experiments 1 and 2 (in Experiment 3, the comparison cannot be made because different types of stimulants were used in the stimulant and stimulant coated applications). Feed intakes of fish

fed the stimulant coated diet were significantly greater than those of the control diet for experiments 1 and 2. There were no significant differences in feed consumption in experiment 3. Growth followed the same pattern as feed intake (data not shown). In each experiment, feed wastage of the stimulant coated was significantly lower than that of the control diet (Figure 2). In experiment 2 wastage of the stimulant coated diet was significantly lower than that of the stimulant diet. In experiment 1, wastage of the stimulant diet was also significantly lower than that of the control diet.

The remaining data are from experiment 1 only, since these results provide a good example of the fishes' response to the feeding stimulants. Results were qualitatively similar in experiments 2 and 3.

Differences in feed intake (Figure 3) were observed after 5 days of feeding and were maintained throughout the experiment. The intakes of all diets were most rapid in the first minute of feeding and slowed as feeding progressed (Figure 4). Differences in feed intake were observed after one minute. In Experiment 1 days 6-10, fish fed the stimulant coated diet ate more in the first minute than the total amount eaten (in 4 minutes) by the control group. After two minutes, consumption by fish fed the stimulant coated diet was greater than the total intake of the fish fed the stimulant diet. Similarly, fish fed the stimulant diet ate more in 2 minutes than the total eaten by the control fish.

Feed wastage is shown in Figure 5. Wastage was highest at the beginning of the trial and declined as the experiment progressed. Five days prior to the time shown in the figure, all diets were rejected. Fish eagerly mouthed the pellets, then spat them out. Figure 6 illustrates wastage in each minute of feeding. Feed wastage was minimal in the first minute of feeding and increased in the final minutes of feeding as satiation was approached. Wastage of the control diet occurred early and gradually, and became noticeable by the second minute. In contrast, wastage of the diets containing the stimulants was not notable until the third minute, after which wastage increased rapidly.

### Discussion

The differences observed in feed intake occurred early, in the first minute of feeding, and in the first five days of the experiment. The fact that the differences occurred so

## Efficacy of Feeding Attractants

quickly indicate that they were a result of the flavor, rather than the nutritional value, of the diet. The visual signs of feeding enthusiasm such as rapid swimming and splashing are often used as qualitative indicators of fish hunger, and these indicators are effectively quantified by measuring short-term feeding rate.

The high feed wastage in experiment 1 can be explained by the fact that all the diets were initially rejected by the fish. The decline in feed wastage that occurred as the experiment progressed is likely a result of the fish gradually becoming accustomed to the diets. The person feeding the fish also adapted, and became better at determining a feeding rate that would maximize opportunity for feeding and minimize feed wastage. The data are therefore subjective and should be interpreted with caution. Wastage data, however, can be a good supplementary, relative measure of diet palatability since the results were qualitatively similar in three experiments. Moreover, given the problems that feed wastage can cause (lost revenue, poorer water quality and greater effluent), the effects of feeding stimulants on this parameter should be given due consideration.

The timing of feed wastage (Figure 6) confirms that the fish were feeding to satiation, and provides a clue concerning the reason for differences in feed wastage. By the second minute of feeding the fish fed the control diet wasted a notable proportion of the ration even though they were still actively feeding. This made it difficult to determine a feeding rate which would meet the apparent demand of the fish without excessive wastage. In contrast when feeding stimulants were used, feed wastage did not become noticeable until the third minute of feeding, when the rate of feed intake was also low.

Coating the pellet with the stimulant seemed to improve its effectiveness. Coating results in higher concentrations of the substances being exposed to the fish, when they disperse through the water and when the feed is grasped by the fish. Further work should be done to determine if using doses higher than those used in this study would improve the feeding response.

The timing of feed intake and feed consumption have implications for feed management which are illustrated in Figure 7. In many rearing systems, feed wastage is difficult to observe in a timely manner, which presents a dilemma for the fish culturist. Although it is desirable to feed the maximum ration the fish will take, when one

nears this feeding level, (i.e. satiation) wastage increases. The use of feeding stimulants may aid the culturist with this problem because not only does wastage tend to be lower for more palatable diets, but a greater proportion of the fishes' total ration is eaten before wastage becomes notable. What this means is that if the fish culturist wishes to minimize feed wastage and ceases feeding prematurely, before complete satiation, there may be less production loss when feeding stimulants are used because palatable diets are eaten more quickly. Alternatively, if the fish culturist is able to use wastage as an indicator of satiation, the improvement of the diet flavor ensures that the fish will be fed more of their ration since wastage of palatable diets tends to occur later in feeding, at a point closer to satiation. It is important to mention that in this study, all the fish had access to feed. In many fish culture situations (i.e. high stocking densities) crowding may prevent all fish from feeding at the same time, and therefore the patterns of feed intake and wastage (and hence one's conclusions) could differ from those of this study.

Perhaps the most promising aspect of the feeding stimulants is that the benefits appear to be long-term. Initially, it was thought that fish fed the less palatable diet would initially eat less and compensate in future feeding sessions; this did not occur. Therefore, feeding stimulants can be another useful tool for the fish culturist to improve the delivery of feed to one's fish.

### Acknowledgments

This study was supported by a grant from Biozyme Systems Ltd., Vancouver, British Columbia. The authors thank Carol MacMillan for technical assistance and Dr. Dave Higgs for his comments on the manuscript.

### References

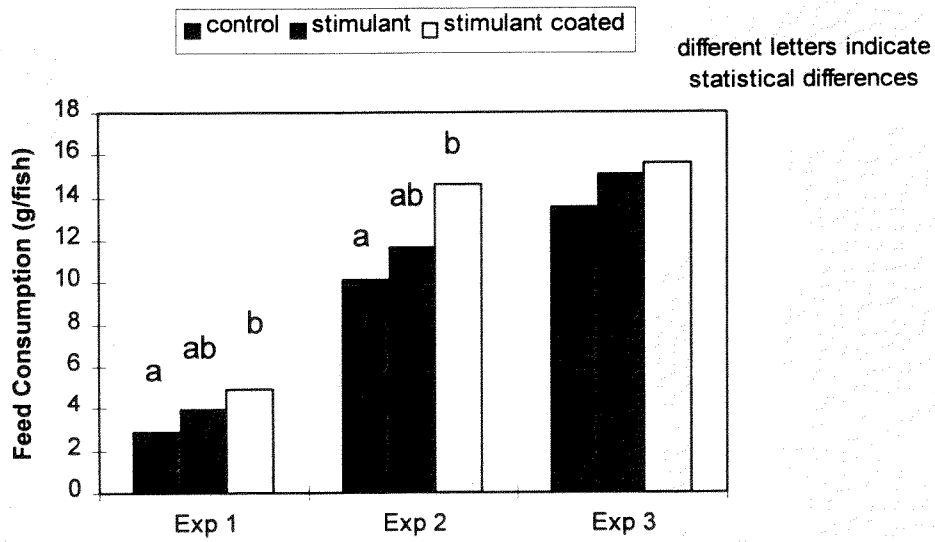
- Adron, J.W. and A.M. Mackie. 1978. Studies on the chemical nature of feeding stimulants for rainbow trout, *Salmo gairdneri*. *Journal of Fish Biology* 12: 303-10.
- Appelbaum, S., J.W. Adron, S.G. George, A.M. Mackie, and B.J. Pirie. 1983. On the development of the olfactory and the gustatory organs of the dover sole, *Solea solea*, during metamorphosis. *Journal of the Marine Biological Association of the U.K.* 63: 97-108.

- Carr, W.E.S. and T.B. Chaney. 1976. Chemical stimulation of feeding behavior in the pinfish, *Lagodon rhomboides*: characterization and identification of stimulatory substances extracted from shrimp. *Comparative Biochemistry and Physiology* 54A: 437-441.
- Jones, K.A. 1989. The palatability of amino acids and related compounds to rainbow trout, *Salmo gairdneri*. *Journal of Fish Biology* 34: 149-160.
- Rumsey, G.L. 1986. Chemical control of feed intake in fishes. *Cornell Nutrition Conference* pp. 100-110.
- Teskeredzic, Z., D.A. Higgs, B.S. Dosanjh, J.R. McBride, R.W. Hardy, R.M. Beames, J.D. Jones, M. Simell, T. Vaara, R.B. Bridges. 1995. Assessment of undephytinized and dephytinized rapeseed protein concentrate as sources of dietary protein for juvenile rainbow trout (*Oncorhynchus mykiss*). *Aquaculture* 131: 261-277.
- Toften H., E.H. Jorgensen and M. Jobling. 1995. The study of feeding preferences using radiography; oxytetracycline as a feeding deterrent and squid extracts as a stimulant in diets for Atlantic salmon. *Aquaculture Nutrition* 1: 145-149.

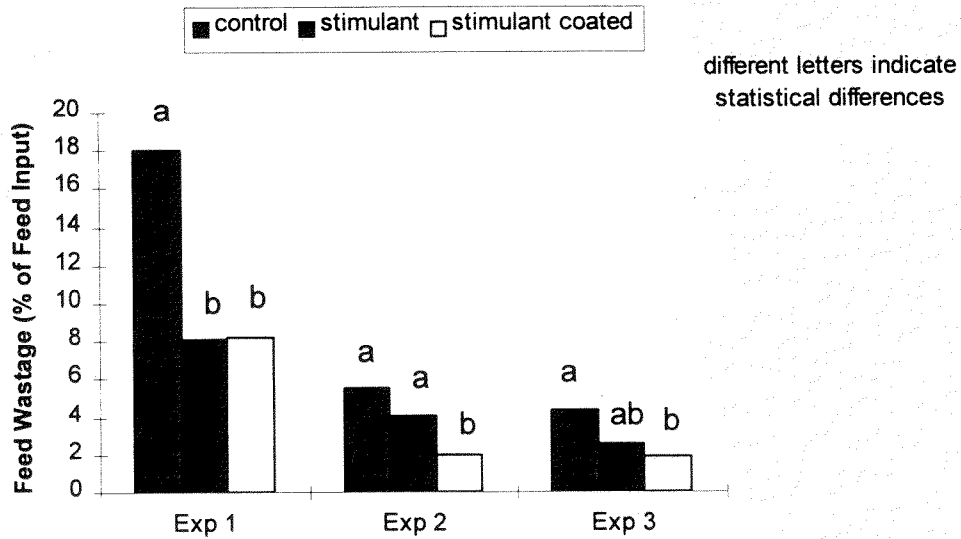


## Efficacy of Feeding Attractants

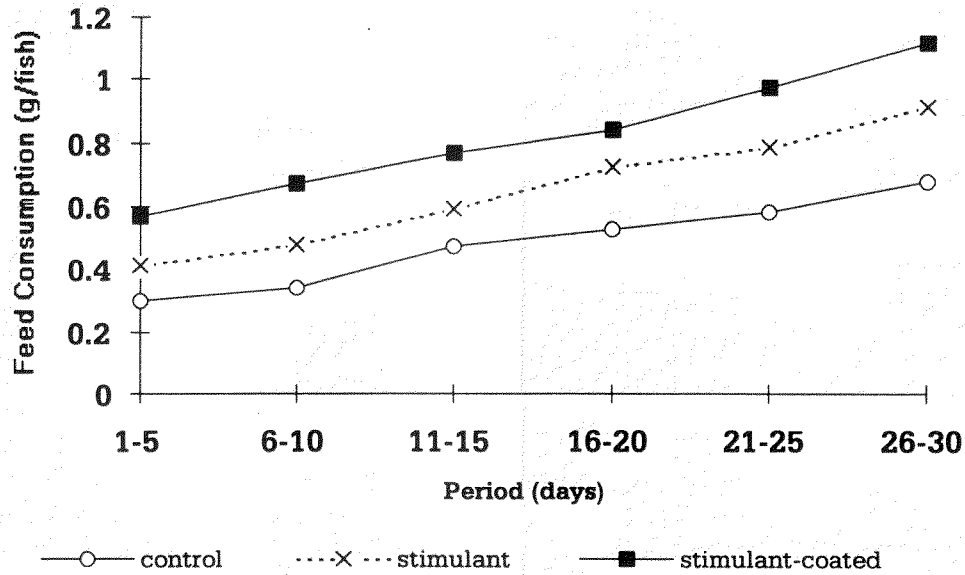
**Figure 1. Feed Consumption in Experiments 1-3**



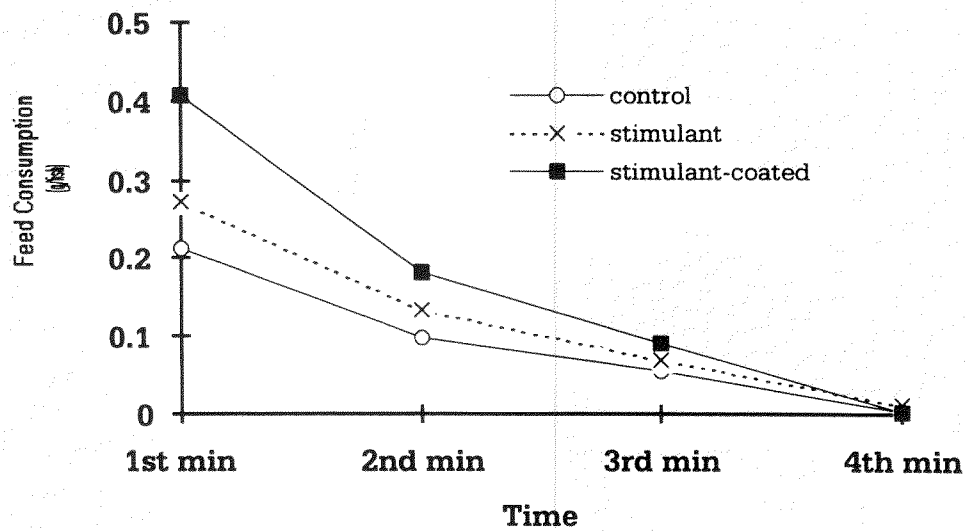
**Figure 2. Feed Wastage in Experiments 1-3**



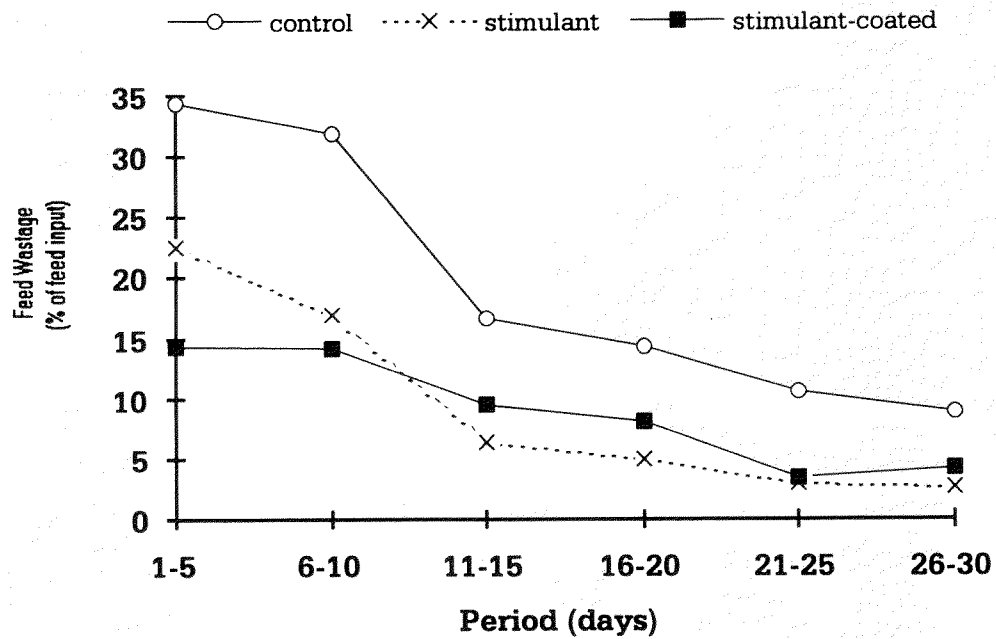
**Figure 3. Feed Consumption in 5-Day Periods by Fish in Experiment 1 (corrected for uneaten feed)**



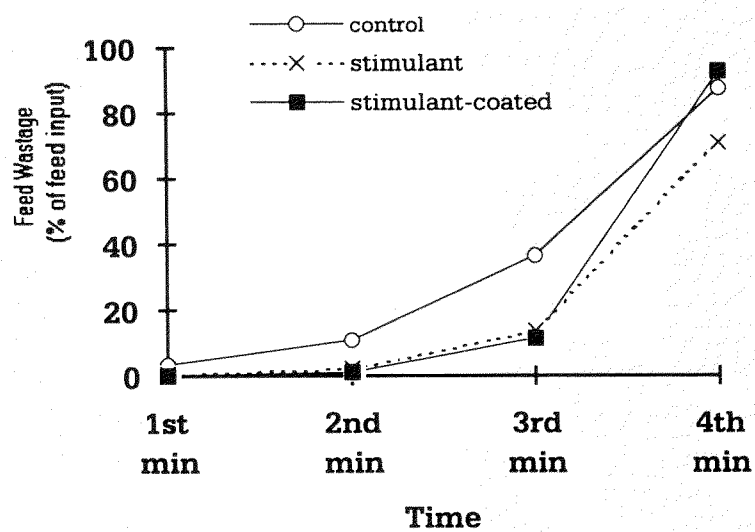
**Figure 4. Feed Consumption Rates of Fish in Experiment 1, Days 6-10 (corrected for uneaten feed)**



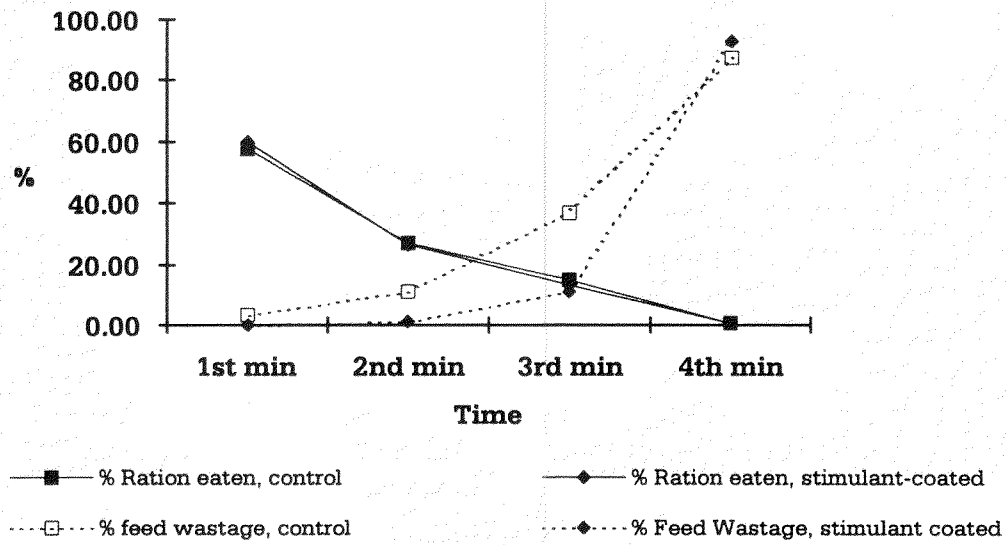
**Figure 5. Feed Wastage in Experiment 1**



**Figure 6. Timing of Feed Wastage by Fish in Experiment 1, Days 6-10**



**Figure 7. Timing of Feed Consumption vs. Feed Wastage  
Experiment 1, Days 6-10**



# Development of Open Formula Diets and New Feeding Strategies: A Progress Report

Ann Gannam

*Abernathy Salmon Culture Technology Center, Longview, WA 98632*

**Abstract** — As more threatened and endangered stocks are moved into a hatchery rearing program, development of life stages diets for the fry and juvenile fish raised in hatcheries is becoming a critical area for research. The new diets would be better adapted to the fish at their various stages of development. Research has been initiated to investigate feeds and feeding practices to help resolve the predominant problems of precocity, fish health, residualism, size distribution at release and other management issues related to reduced out migration of juveniles and diminished adult returns.

Studies already completed include: testing of a low fat Abernathy dry diet, developing open formula semi-moist feeds using Abernathy's new Wenger X85 cooker-extruder and adding immunomodulators to the diet, testing the resulting feeds at the Abernathy Salmon Culture Technology Center and, cooperatively, at the Columbia River Research Laboratory, Biological Resources Division, U.S. Geological Survey. A pilot study has been started at the Dworshak National Fish Hatchery in cooperation with the Columbia River Research Laboratory to evaluate feeding regimes.

## Introduction

Nutrition research is essential when hatchery fish will be used for supplementation of weak wild and naturally spawning fish populations. In supplementation/enhancement programs, diets are needed that will maintain the health of the fish as well as produce fish that are "wild" in appearance and composition. Current hatchery production fish will also benefit from new diets. Habitat destruction has brought about the realization that there will be a greater dependance on hatchery raised stocks to replenish fish runs in the Columbia Basin and elsewhere. Open formula diets in these rearing programs are also important. In open formula diets the ingredients are known and can be specified. In addition, because the formulas are open, they can be monitored through quality control programs. Thus there is a need to develop new open formula diets for use in hatchery production.

A number of studies have been completed at the Abernathy Salmon Culture Technology Center (SCTC) in the past three years concerning the nutrition of hatchery raised salmon. The studies included testing a practical formulation for a low fat diet, feeding open formula semi-moist feeds developed using the cooker-extruder and trying several glucan containing compounds in the diet to evaluate their immunomodulator activity in fall chinook. A study concerning feeds and feeding regimes that was initiated by the Dworshak Fisheries Resource Office (FRO) in 1997 in cooperation with the Columbia River Research Laboratory, Biological Resources Division, U.S. Geological Survey and Abernathy SCTC is ongoing.

Levels of fat as well as composition of the fat are important. Dupree et al. (1979) reported that as the lipid content of the diet for channel catfish increased from 0% to 20%, the whole fish lipid levels increased from 3.8% to

13.2%. In a study using turbot, Caceres-Martinez et al. (1984) saw a negative effect in the fish tissue of excess dietary lipids. At the lowest protein level, 37.5%, the tissue lipid deposition increased as the lipid level in the feed increased from 10% to 20%. Several investigators have shown that the fatty acid content of the lipids used in fish diets are important for good growth (Stickney and Andrews 1972, Heck and Calbert 1977, Farkas, et al. 1977, Stickney et al. 1984).

The importance of fatty acids in the correct proportions in the diets is demonstrated in the study done by Lewis et al. (1985). Catfish fed various combinations of stearic, oleic, linoleic, and linolenic acids performed poorly. Castell et al. (1994) also showed the importance certain fatty acids in fish feeds. They found that arachidonic acid (20:4n-6) may be an essential fatty acid for juvenile turbot. Arachidonic acid is not given much attention in fish diets, however, Bell et al. (1994) found in the 10 freshwater invertebrates (common prey for salmon) he analyzed for fatty acid composition that there were higher levels of 18:2n-6, 18:3n-3, 20:4n-6 and 20:5n-3 and less 22:6n-3 than found in commercial diets used in smolt production. In a study done by Lie et al. (1986), three different fats were tested in diets for cod. They used cod liver oil, Greenland halibut oil and peanut oil. The study indicated that the type and amount of fat used influenced fat deposition in the liver.

The liver is an indicator of the quality of the diet is for the fish. Fowler and Wood (1966) tested different supplemental dietary fats on chinook salmon fingerlings. The investigators found the when chinook salmon were fed a meat diet or an all-meat diet supplemented with hard animal fat as the principal lipid source fatty liver and degenerative changes of the spleen and hematopoietic part

of the kidney occurred. Use of a vegetable oil instead of animal fat prevented these abnormalities.

Carbohydrate levels in the diet of salmonids are an important consideration, especially now that extrusion of feeds makes the carbohydrate included in the diet more digestible. Feeding high levels of digestible carbohydrate to salmon has resulted in increased liver size and glycogen content which is proportional to the carbohydrate fed (Wilson 1994). Starch inclusion in the diet of Atlantic salmon higher than 22% had negative effects on growth and feed utilization (Hemre et al. 1995). Inclusion of starch above 9% resulted in decreased starch digestibility. Ashley (1972) and Roberts and Bullock (1989) discuss the pathologies of excessive carbohydrates in the diet of fish.

To examine the fat and carbohydrate questions, two diet studies were done, one to test a practical low fat diet and the other to determine the appropriate level of carbohydrate to incorporate into the open formula semi-moist feed.

Glucans administered by intraperitoneal injection or as a bath have been used to enhance the nonspecific immune response in Atlantic salmon, *Salmo salar*, coho, *Oncorhynchus kisutch*, channel catfish, *Ictalurus punctatus*, and brook trout, *Salvelinus fontinalis*, (Robertsen et al. 1990, Nikl et al. 1991, Chen and Ainsworth 1992, Anderson and Siwicki 1994). Engstad et al. (1992) and Jorgensen et al. (1993) found that the lysozyme levels in the blood of Atlantic salmon and rainbow trout, *Oncorhynchus macaw*s, respectively, increased after an intraperitoneal (i.p.) injection of a glucan from *Saccharomyces cerevisiae*. Matsuyama et al. (1992) also found an increase in serum lysozyme when yellowtail *Seriola quinqueradiata* were injected i.p. with  $\beta$ -1,3-glucans derived from *Schizophyllum commune* and *Sclerotium glaucum*.

Little work examining the effects of orally administered glucans on the fish's immune response has been done. Raa et al. (1992) achieved positive results feeding Macrogard, a  $\beta$ -1,3/1,6-glucan from yeast, to Atlantic salmon. Fish fed the glucan at 1 g/kg dry feed sustained a survival of approximately 60%. Whereas the fish fed the control diet had a 20% survival after a challenge with *Vibrio salmonicida*. Survivals were better (~85%) when the glucan fed fish were exposed to *V. anguillarum*. In addition, Siwicki et al. (1994) fed several

immunostimulant preparations mixed into semipurified diets to rainbow trout. In the fish fed the immunostimulants there was an increase in oxidative radical release, myeloperoxidase activity, phagocytic indexes and potential killing activities of phagocytic cells including neutrophils.

The present trial examined the effect of orally administered immunostimulants on the nonspecific immune responses of fall chinook salmon juveniles. Lysozyme increase was the response monitored.

The Draft Snake River Salmon Recovery Plan (SRSRP) and the Biological Opinion from the National Marine Fisheries Service (NMFS) on Hatchery Operations for 1996-1999 calls for hatchery steelhead to be released between 170-220 mm (TL) in order to minimize residualization. Steelhead larger than 170 mm experience more complete parr-smolt transformation and are therefore more likely to actively migrate. Fish larger than 220 mm are more prone to residualize (Partridge 1985, 1986; Cannamela 1992). In addition, steelhead greater than 250 mm may be more capable of predation (Cannamela 1993). Those steelhead that fail to emigrate, both large and small, may negatively impact sensitive fish species such as the endangered Snake River fall chinook, spring/summer chinook, or the proposed threatened wild Snake River steelhead through displacement, competition for food, and/or behavioral influences (Viola and Schuck 1995). Juvenile hatchery steelhead have been collected in several tributary streams below Dworshak NFH after smolts were released in 1994, 1995, and 1996. The primary objective of the SRSRP recommendation is to reduce the potential impacts of various state and federal hatchery programs on listed fish species by attempting to minimize the number of non-migrating hatchery summer steelhead smolts released into the Snake River basin.

The goal of this project is to significantly decrease the number of summer steelhead *Oncorhynchus mykiss* produced at Dworshak National Fish Hatchery that residualize in the river and fail to migrate to the ocean as smolts after being released. Diets and an altered feeding regime are being tested to accomplish the goal.

## Materials and Methods

### Lipid level study

## Development of Open Formula Diets and New Feeding Strategies

The fish used in the lipid level experiment were fall chinook salmon (*Oncorhynchus tshawytscha*) fingerlings raised from eggs at the Abernathy Salmon Culture Technology Center (SCTC). These fish, average weight 7.4 g, were stocked into 700 liter circular fiberglass tanks, 250 fish per tank. Four tanks were randomly assigned to each treatment. Well water (12°C) was used at 19 liters/minute.

The treatments consisted of the basic Abernathy diet with different levels of lipid used in the formulation. The levels of lipid used were 9.0%, 11.4%, 19.9% and the control, 15.7% (Table 1). Proximate analysis (AOAC 1990) of the feed ingredients for the experimental diets were determined and the diets formulated. A small compaction-type pellet mill (California Pellet Mill, San Francisco, CA), without steam conditioning, was used to prepare the diets. Feed was made at the start of the experiment and stored at room temperature.

The fish were fed by hand four times a day, five days a week for 10 weeks. On the weekends the fish were fed with automatic feeders four times a day. The amounts of feed fed were calculated by the method of Buterbaugh and Willoughby (1967) based on a hatchery constant of nine. The fish were weighed every two weeks and the amount feed was adjusted accordingly.

The data reported for the feeding trial included average weight gain, gross feed conversion, hepatosomatic indexes as well as liver glycogen and liver triglyceride levels. Liver analyses were done by Biotech Research and Consulting, Inc., Corvallis, OR. Proximate analysis was done on the fish and the feed. The data were analyzed using the one-way analysis of variance to determine if there were differences between treatments ( $P < 0.05$ ). Where differences were found, the treatments that were different from each other were determined by the Student-Newman-Keuls method ( $P < 0.05$ ) (Ostle and Mensing 1975).

### *Semi-moist diet study*

In the open formula semi-moist diet study the fish used were fall chinook salmon (*Oncorhynchus tshawytscha*) fingerlings, Abernathy SCTC stock. These fish, average weight 5.5 g, were stocked into 700 liter circular fiberglass tanks, 250 fish per tank. Four tanks were randomly assigned to each treatment. Well water (12°C) was used at 19 liters/minute.

The treatments consisted of the three semi-moist formulations and the Abernathy Dry diet as a control (Table 2). The amount of wheat product in the diet was varied to determine the least amount that could be used and still produce a pellet with desirable characteristics. Proximate analysis (AOAC 1990) of the feed ingredients for the experimental diets were determined and the diets formulated. A Wenger X85 single screw cooker-extruder was used to make the feeds. The diets were made at the start of the experiment and stored at room temperature in plastic containers. The fish were fed seven days a week with automatic feeders four times a day for 10 weeks. The amounts of feed fed were calculated by the method of Buterbaugh and Willoughby (1967) based on a hatchery constant of nine. The fish were weighed every two weeks and the amount feed was adjusted accordingly. The quantities of feed fed to the different treatments was also corrected for moisture in the feed.

The data reported for the feeding trial included average weight gain, gross feed conversion, hepatosomatic indexes as well as liver glycogen and liver triglyceride levels. Liver analyses were done by Biotech Research and Consulting, Inc., Corvallis, OR. The data was analyzed using the one-way analysis of variance to determine if there are differences between treatments ( $P < 0.05$ ). Where differences were found, the treatments that were different from each other were determined by the Student-Newman-Keuls method ( $P < 0.05$ ) (Ostle and Mensing 1975).

### *Glucan diet study*

For the glucan feeding trial all compounds were added to a modified Abernathy diet (Table 3). The respective percentages of the compounds were substituted for mill run (Table 4). The diets were made at the Abernathy SCTC. The dosage period for each compound was 14 days.

Fall chinook salmon raised at the Abernathy SCTC, average weight 5.1 g, were tested for initial serum lysozyme, mucus lysozyme from the skin, nares and intestinal wall, and for gill sodium, potassium ATPase. The fish were then transported to the Columbia River Research Laboratory. At the facility stock fish were held in heated well water in a 1400 liter circular tank at 6 liters/minute flow, equipped with air stones, and kept at 12-13°C. Fish stocking densities did not exceed guidelines found for fall chinook salmon (Piper et al. 1982). After a one week acclimation period, groups of 30

fish were weighed and sampled for skin mucus lysozyme, and transferred to 228 liter circular tanks, in 12-13°C well water, with flows of 6 liters/minute. Fish will be kept on natural photoperiod. Fish were randomly assigned to the eight experimental tanks, with duplicates of the following treatment groups: two control tanks fed Abernathy Control Diet, two tanks fed Levucell SB, two tanks fed ALGAMAC-2000 (*Schizochytrium*), and two tanks fed VitaStim (VST).

Levucell is a live cell yeast preparation and is reported to compete with pathogenic bacteria in the intestine by attaching to them, preventing their adhesion to the digestive tract and by inhibiting pathogenic toxins (manufacturers information). ALGAMAC, a freeze dried preparation of the macro algae, *Schizochytrium*, contains intact cell walls (manufacturer information). VitaStim has been shown to improve survival of fish after challenge with *Aeromonas salmonicida* when used as a vaccine injection adjuvant (Nikl et al. 1991). Dosages are in Table 4.

At the start of the feeding study, 30 fish from the stock tank will be sampled for serum lysozyme, mucus lysozyme from the skin, nares and intestinal wall, and for gill sodium, potassium ATPase. All skin mucus samples for lysozymes were collected from the lateral line area above the vent. Nares mucus were obtained by inserting the loop into the nares and removing the loop after turning it ½ turn. Intestinal mucus was collected by inserting the loop into the intestine to the first taper of the loop handle and drawing it out while gently scraping the intestinal wall by providing pressure on the outside of the fish. Blood was obtained by severing the caudal peduncle and collected in Microtainer brand serum separation tubes (Becton Dickinson & Co. Rutherford, N. J. 07070). All samples were frozen until analyzed. For all sampling, fish were anesthetized using a 40 mg/L MS-222 solution buffered with sodium bicarbonate. The lysozyme analyses were done by the Columbia River Research Laboratory, USGS, Cook, WA. The methods of collection and analysis were described by Schrock (1994).

After a two week feeding regime at 2% body weight, the fish fasted for 24 hours, then anesthetized with 40 mg/liter MS-222, weighed and sampled for skin mucus lysozyme. After 20-30 minute recovery period the fish were challenged. The fish were not fed on the day of challenge, but feeding resumed the following day, and all groups were fed Abernathy Control Diet at 2% body weight/day.

### Disease challenge

Fish were challenged with *Vibrio anguillarum*. The culture was prepared from a lyophilized preparation, L8-173, 9-14-95, from Chris Banner at Oregon State University. A vial of the culture was added to 1 liter of sterile 1% NaCl tryptic soy broth, and allowed to grow until the optical density reading at 405nm is > 0.800. Verification of the presence of viable *V. anguillarum* was done by culturing of the broth on 1% NaCl tryptic soy agar plates at the time of the last tank challenge which should result in the growth of cream colored, round, raised colonies. Fish in the recovery buckets containing 13 liter heated well water were challenged with 10 ml of the inoculated broth for 1 hour under constant aeration. At exactly 1 hour, fish were returned to their experimental tanks, and observed for signs of infection. At the time mortality reached 50% the remaining fish in each tank were anesthetized and sampled for the following: serum lysozyme, mucus lysozyme from the skin, nares and intestine, and the kidney, spleen, and liver.

### Replicate experiment

The experiment was repeated one month later to determine the effects of the level of physiological development and smoltification on the lysozyme levels, and the response of the fish to the special diets.

For statistical analysis, analysis of variance was used to determine if there was a difference between treatments at the 0.05 level of significance. Where the treatments were significantly different, the Student-Newman-Keuls method was used to ascertain where the differences occurred (Ostle and Mensing 1975).

### Results and Discussion

For the lipid level study, weight gain and gross feed conversion were not significantly different ( $P>0.05$ ). Proximate composition of the fish fed the various levels of lipid in the diet did show some significant differences (Table 5). The percent lipid, on a wet weight basis, in the fish fed the low lipid diet was significantly less than ( $P<0.05$ ) the lipid in the fish fed the high lipid and control diets. Not surprisingly, moisture levels were also significantly different with the low fat fed fish having the highest moisture. In cultured fish, moisture is inversely related to lipid levels (Shearer 1994). The hepatosomatic indexes were not significantly different between groups ( $P>0.05$ ) (Table 5). Smoltification and precocity in



salmonids have been shown to be affected by stored fat (Ogata and Konno 1986, Silverstein et al. 1997). When examining the whole body composition of the fish in this feeding trial, on a wet weight basis, (Shearer 1994), the lipid composition of the fish was decreased when they were maintained on a reduced fat diet. No differences were seen in glycogen or triglyceride content of the livers (Table 6). There were also no differences in the hepatosomatic indexes.

The fish in the semi-moist feeding trial did not show a significant difference in weight gain or feed conversion ( $P>0.05$ ). Analysis of the proximate composition of the fish is not complete. Differences in the hepatosomatic indexes could not be determined because of too much variation within treatments. The triglyceride levels in the livers were not significantly different ( $P>0.05$ ) however the glycogen levels were ( $P<0.05$ ). The liver glycogen levels of the fish follow the carbohydrate levels found in the various diets (Table 2) with an exception. The control diet has a carbohydrate level similar to the semi-moist #3 but the control was not cooked in the extruder and its carbohydrate was not as available. Excessive carbohydrate intake will result in excessive glycogen deposits in the liver (Ashley 1972, Roberts and Bullock 1989)

In the glucan diet trial, the first challenge study, Levucell showed a significantly longer time ( $P<0.05$ ) to 50% mortality (163 hr) after the bath challenge with *Vibrio anguillarum*. Time to 50% mortality for the *Schizochytrium* (123 hr) and VitaStim (VST) (120 hr) groups was significantly longer ( $P<0.05$ ). Levucell's main action is to bind to gram-negative bacteria in the gut and remove them from the system. In addition, to have a positive action the Levucell must be alive. This yeast could have been killed during the storage period between the two challenge studies.

The fish, in the first study, fed the VST and *Schizochytrium* diet were significantly larger ( $P<0.05$ ) than the fish fed the control diet and the VST fed fish were significantly larger ( $P<0.05$ ) than the Levucell fed fish. The differences in percent weight gain seen in the two studies could be explained by the stage of smolt (Table 8).

Skin mucus lysozyme concentrations did not differ significantly ( $P>0.05$ ) among the feed groups at any time in the first challenge study and were low ( $<45 \text{ ug ml}^{-1}$  HEWL) during the entire experiment. Levels increased from a mean of  $24 \text{ ug ml}^{-1}$  HEWL ( $n=241$ ,  $SE=1$ ) at the initial sampling to  $34$  ( $n=233$ ,  $SE=2$ ) in all groups after

the two week feeding period. Levels in survivors at the time of 50% mortality were extremely low with a mean of  $14$  ( $n=121$ ,  $SE=2$ ). The mean nares mucus lysozyme level of  $256$  ( $n=119$ ,  $SE=21$ ) and vent mucus lysozyme  $162$  ( $n=120$ ,  $SE=5$ ) levels were also low compared to other species (Schrock 1994). The differences among groups were not significant (Table 9).

The disease challenge in the second study was much less severe as evidenced by a longer time to onset of mortality in all groups and no group had reached 50% mortality at the termination of the study. The preparation of the broth was exactly the same but a different vial of lyophilized pathogen was used. Lysozyme levels in the second study never exceeded 60. The initial mean skin lysozyme concentration  $21$  ( $n=232$ ,  $SE=1$ ) and the concentration after the feeding period,  $20$ , did not differ ( $n=237$ ,  $SE=2$ ). The mean concentration at the time of termination was  $43$  ( $n=118$ ,  $SE=2$ ). There were no significant differences among groups during the experiment. The mean nares mucus lysozyme  $255$  ( $n=120$ ,  $SE=13$ ) and mean vent mucus lysozyme  $194$  ( $n=119$ ,  $SE=6$ ) were comparable to the levels found in the first study (Table 9).

Other experiments testing the effects of oral administration of glucans have resulted in contradictory results (Siwicki et al. 1994). The effects of size, age and physiological development need to be examined. In the present study the fish may have had immune system suppression associated with smoltification (Maule et al. 1987, 1993). In addition, the increase in specific components of the immune response observed in other fish fed immunostimulants do not necessarily increase survival in a challenge situation (Duncan and Klesius 1996). Contradictions remain between the studies based on mode of administration, dose and challenge pathogen. Each immunomodulator preparation must be considered individually by mode of application, dose and anticipated pathogen to evaluate the potential of using immunomodulators to improve disease resistance in fish.

## References

- Anderson, D. P. and A. K. Siwicki. 1994. Duration of protection against *Aeromonas salmonicida* in brook trout immunostimulated with glucan or chitosan by injection or immersion. *Progressive Fish-Culturist* 56:258-261.
- Ashley, L. M. 1972. Nutritional pathology. In, *Fish nutrition*, J. Halver, ed. pp. 490-492. Academic Press, New York.
- Association of Official Analytical Chemists. 1990. Official methods of analysis of the Association of Official Analytical Chemists, 15th ed. S. Williams (Editor). Association of Official Analytical Chemists, Arlington, Va. 1141 pp.
- Bell, J. G., C. Ghioni and J. R. Sargent. 1994. Fatty acid compositions of 10 freshwater invertebrates which are natural food organisms of Atlantic salmon parr (*Salmo salar*): a comparison with commercial diets. *Aquaculture* 128:301-313.
- Buterbaugh, G. L. and H. Willoughby. 1967. A feeding guide for brook, brown, and rainbow trout. *Progressive Fish-Culturist* 29: 210-215.
- Caceres-Martin, C. M. Cadena-Roa and R. Metailler. 1984. Nutritional requirements of turbot (*Scophthalmus maximus*): I. A preliminary study of protein and lipid utilization. *Journal of the World Mariculture Society* 15:191-202.
- Cannamela, D.A. 1992. Potential impacts of releases of hatchery steelhead trout "smolts" on wild and natural juvenile chinook and sockeye salmon. A white paper. Copies available from Idaho Department of Fish and Game, Boise, Idaho. 36p.
- Cannamela, D.A. 1993. Hatchery steelhead smolt predation of wild and natural juvenile chinook salmon fry in the upper Salmon River, Idaho. A white paper. Copies available from Idaho Department of Fish and Game, Boise, Idaho. 23p.
- Castell, J. D., J. G. Bell, D. R. Tocher and J. R. Sargent. 1994. Effects of purified diets containing different combinations of arachidonic and docosahexaenoic acid on survival, growth and fatty acid composition of juvenile turbot (*Scophthalmus maximus*). *Aquaculture* 128: 315-333.
- Chen, D. and A. J. Ainsworth. 1992. Glucan administration potentiates immune defense mechanisms of channel catfish, *Ictalurus punctatus* Rafinesque. *Journal of Fish Diseases* 15:295-304.
- Duncan, P. L. and P. H. Klesius. 1996. Dietary immunostimulants enhance non-specific immune responses in channel catfish but not resistance to *Edwardsiella ictaluri*. *Journal of Aquatic Animal Health* 8: 241-248.
- Dupree, H., E. Gauglitz, Jr., A. Hall and C. Houle. 1979. Effects of dietary lipids on the growth and acceptability (flavor) of channel catfish (*Ictalurus punctatus*). *Proc. World Symp. on Finfish Nutrition and Fishfeed Technology*, Hamburg 2:87-103.
- Engstad, R. E., B. Robertsen and E. Frivold. 1992. Yeast glucan induces increase in lysozyme and complement-mediated haemolytic activity in Atlantic salmon blood. *Fish & Shellfish Immunology* 2:287-297.
- Fowler, L. G. and E. M. Wood. 1966. Effect of type of supplemental dietary fat on chinook salmon fingerlings. *Progressive Fish Culturist*. 26:123-127.
- Heck, N. E. and H. E. Calbert. 1977. Use of animal fat in formulated diets for yellow perch. *Proceedings of the eighth annual meeting world Mariculture Society*; 1977 January 9-13; San Jose, Costa Rica. c1977:787-791.
- Hemre, G-I., K. Sanders, O. Lie, O. Torrissen, R. Waagbo. 1995. Carbohydrate nutrition in Atlantic salmon, *Salmo salar* L.: growth and feed utilization. *Aquaculture Research* 26:149-154.
- Jorgensen, J. B., G. J. E. Sharp, C. J. Secombest, and B. Robertsen. 1993. Effect of a yeast-cell-wall glucan on the bactericidal activity of rainbow trout macrophages. *Fish & Immunology* 3:267-277.
- Landolt, M. L. 1989. The relationship between diet and the immune response of fish. *Aquaculture* 79: 193-206.
- Lewis, D. H., J. E. Marks and R. R. Stickney. 1985. Degenerative myopathy in channel catfish, *Ictalurus punctatus* (Rafinesque), maintained on rations

# Development of Open Formula Diets and New Feeding Strategies

- containing purified fatty acids. *Journal of Fish Diseases* 8:563-565.
- Li, M. H. and E. H. Robinson. 1996. Comparison of chelated zinc and zinc sulfate as zinc sources for growth and bone mineralization of channel catfish (*Ictalurus punctatus*) fed practical diets. *Aquaculture* 146:237-243.
- Lie, O., E. Lied, and G. Lambertsen. 1986. Liver retention of fat and of fatty acids in cod (*Gadus morhua*) fed different oils. *Aquaculture* 59:187-196.
- Matsuyama, H., R. E. P. Mangindaan, T. Yano. 1992. Protective effect of schizophyllan and scleroglucan against *Streptococcus* sp. infection in yellowtail (*Seriola quinqueradiata*). *Aquaculture* 101:197-203.
- Maule, A. G., C. B. Schreck and S. L. Kaattari. 1987. Changes in the immune system of coho salmon (*Oncorhynchus kisutch*) during the parr-smolt transformation and after implantation of cortisol. *Canadian Journal of Fisheries and Aquatic Science* 44: 161-166.
- Maule, A. G., C. B. Schreck and C. Sharpe. 1993. Seasonal changes in cortisol sensitivity and glucocorticoid receptor affinity and number in leukocytes of coho salmon. *Fish Physiology and Biochemistry* 10: 497-506.
- Mazur, C. N., D. A. Higgs, E. Plisetskaya and B. E. March. 1992. Utilization of dietary starch and glucose tolerance in juvenile chinook salmon (*Oncorhynchus tshawytscha*) of different strains in seawater. *Fish Physiology and Biochemistry* 10:303-313.
- Nikl, L., L. J. Albright, T. P. T. Evelyn. 1991. Influence of seven immunostimulants on the immune response of coho salmon to *Aeromonas salmonicida*. *Diseases of Aquatic Organisms* 12:7-12.
- Ogata, H. and S. Konno. 1986. Growth response and smolt production of 1 year cherry salmon fed diets having different protein and lipid levels. *Bull. Japn. Soc. Sci. Fish.* 52:313-318.
- Ostle, B. And R. Mensing. 1975. *Statistics in research.* 596 pp. The Iowa State University Press, Ames.
- Partridge, F.E. 1985. Effects of steelhead smolt size on residualism and adult return rates. U.S. Fish and Wildlife Service, Lower Snake River Compensation Plan. Contract No. 14-16-001-83605. Idaho Department of Fish and Game, Boise, Idaho.
- Partridge, F.E. 1986. Effects of steelhead smolt size on residualism and adult return rates. U.S. Fish and Wildlife Service, Lower Snake River Compensation Plan. Contract No. 14-16-001-83605. Idaho Department of Fish and Game, Boise, Idaho.
- Piper, R., I. McElwain, L. Orme, J. McCraren, L. Fowler and J. Leonard. 1982. *Fish Hatchery Management.* U. S. Department of Interior, Fish and Wildlife Service, Washington, D. C. pp. 517
- Raa, J., G. Roerstad, R. Engstad and B. Robertsen. 1992. The use of immunostimulants to increase resistance of aquatic organisms to microbial infections. In: *Diseases in Asian Aquaculture I.* M. Shariff, R. P. Subasinghe and J. R. Arthur, eds. pp. 39-50. Fish Health Section, Asian Fisheries Society, Manila, Philippines.
- Robertsen, B., G. Roerstad, R. Engstad and J. Raa. 1990. Enhancement of non-specific disease resistance in Atlantic salmon, *Salmo salar* L., by a glucan from *Saccharomyces cerevisiae* cell walls. *Journal of Fish Diseases* 13:391-400.
- Roberts, R. J. and A. M. Bullock. 1989. Nutritional pathology. In, *Fish Nutrition*, 2nd edition, J. Halver, ed. p. 430. Academic Press, Inc., San Diego, New York.
- Schrock, R. M. 1994. Quantifying non-specific disease response in adult and juvenile salmon. In *Proceedings of an International Fish Physiology Symposium*, Physiology Section, American Fisheries Society and the Fish Physiology Association. Vancouver, B. C. July 16-21, 1994.
- Shearer, K. D. 1994. Factors affecting the proximate composition of cultured fishes with emphasis on salmonids. *Aquaculture* 119:63-88.
- Silverstein, J. T., H. Shimma and H. Ogata. 1997. Early maturation in amago salmon: an association with energy storage. *Canadian Journal of Fisheries and Aquatic Sciences* 54:444-451.
- Siwicki, A. J., D. P. Anderson and G. L. Rumsey. 1994. Dietary intake of immunostimulants by rainbow trout affects non-specific immunity and protection against furunculosis. *Veterinary Immunology and Immunopathology* 41:125-139.

Stickney, R. R. and J. W. Andrews. 1972. Effects of dietary lipids on growth, food conversion, lipid and fatty acid composition of channel catfish. *Journal of Nutrition* 102:249-258.

Stickney, R. R., R. B. McGeachin and E. H. Robinson. 1984. Effect of dietary linoleic acid level on growth, food conversion and survival of channel catfish. *Journal World Mariculture Society* 15:186-190.

Viola, A.E. and M.L. Schuck. 1995. A method to reduce the abundance of residual hatchery steelhead in rivers. *N. M. J. Fish Manage.* 15:488-493.

Wilson, R. P. 1994. Utilization of dietary carbohydrate by fish. *Aquaculture* 124:67-80.

## Development of Open Formula Diets and New Feeding Strategies

Table 1. Composition and Proximate analysis of the feed used in the lipid level study

Ingredients	Low Lipid	Medium	High	Control
Anchovy meal	51.90	51.90	51.90	51.90
Feather meal	10.00	10.00	10.00	10.00
Blood meal	2.50	2.50	2.50	2.50
Wheat germ	5.00	5.00	5.00	5.00
Mill run	21.27	19.27	9.27	14.27
Wheat flour	5.00	5.00	5.00	5.00
Vitamins <sup>1</sup>	1.50	1.50	1.50	1.50
Choline	0.58	0.58	0.58	0.58
Ascorbic acid	0.20	0.20	0.20	0.20
Minerals <sup>1</sup>	0.05	0.05	0.05	0.05
Fish oil	2.00	4.00	14.00	9.00
<b>Proximate analysis</b>				
% Protein	52.2	52.5	50.9	51.5
% Lipid	9.0	11.4	19.9	15.4
% Moisture	8.1	7.9	7.0	7.5
% Ash	10.2	10.0	9.7	9.8

<sup>1</sup>The vitamin and mineral premixes supply the following concentrations (mg/Kg diet): choline chloride, 3500; biotin, 0.6; B<sub>12</sub>, 0.6; folic acid, 16.5; inositol, 132; vitamin K, 9.2; niacin, 222; pantothenic acid, 106; pyridoxine, 30.9; riboflavin, 52.9; thiamin, 43.0; vitamin E, 503 IU; vitamin A, 6614 IU; vitamin D, 441 IU; zinc, 75; manganese, 20; copper, 1.5; and iodine, 10.

<sup>2</sup> Fish oil stabilized with 500 mg of liquid ethoxyquin per kg of oil; 4% oil added during pelleting and the remainder as a top dressing.

Table 2. Composition and proximate analysis of the semi-moist feeds

Ingredients	Semi #1	Semi #2	Semi #3	Control
Anchovy meal	46.90	44.00	46.00	45.30
Feather meal	7.50	7.50	10.00	10.00
Blood meal	5.00	5.00	2.50	2.50
Wheat germ	2.50	2.50	5.00	5.00
Mill run	0.828	1.728	8.678	19.278
Wheat flour	5.00	7.00	10.00	5.00
Vitamins <sup>1</sup>	1.50	1.50	1.50	1.50
Choline	0.58	0.58	0.58	0.58
Ascorbic acid (Stay-C)	0.05	0.05	0.05	0.20 <sup>3</sup>
Minerals <sup>1</sup>	0.10	0.10	0.10	0.10
Lignin	0.417	0.417	0.417	0.417
Propylene glycol	2.00	2.00	2.00	-
Sorbate	0.30	0.30	0.30	-
Ca propionate	0.125	0.125	0.125	0.125
Water	14.6	14.6		
Fish oil <sup>2</sup>	12.6	12.6	12.6	10.00
<b>Proximate analysis (dry matter basis)</b>				
% Protein	56.4	53.1	49.0	50.7
% Lipid	20.5	19.5	19.3	16.4
% Ash	10.5	9.9	9.1	10.5
%Carbohydrate	11.0	15.9	21.0	20.8

<sup>1</sup> The vitamin and mineral premixes supply the following concentrations (mg/Kg diet): choline chloride, 3500; biotin, 0.6; B<sub>12</sub>, 0.6; folic acid, 16.5; inositol, 132; vitamin K, 9.2; niacin, 222; pantothenic acid, 106; pyridoxine, 30.9; riboflavin, 52.9; thiamin, 43.0; vitamin E, 503 IU; vitamin A, 6614 IU; vitamin D, 441 IU; zinc, 75; manganese, 20; copper, 1.5; and iodine, 10.

<sup>2</sup> Fish oil stabilized with 500 mg of liquid ethoxyquin per kg of oil; 10% oil was added during extruding and the remainder as a top dressing. For the Abernathy Diet, 4% oil was added before pelleting and the remainder as a top dressing.

<sup>3</sup> Crystalline vitamin C was used in the Abernathy Diet (Control).

Table 3. Approximate formulations and proximate compositions of the glucan diets.

# Development of Open Formula Diets and New Feeding Strategies

Ingredient (%)	Control	Levucell	<i>Schizochytrium</i>	VST
Anchovy meal	50.50	50.50	50.50	50.50
Feather meal	10.00	10.00	10.00	10.00
Blood meal	2.50	2.50	2.50	2.50
Wheat flour	5.00	5.00	5.00	5.00
Mill run	14.27	13.77	13.27	14.17
Wheat germ	5.00	5.00	5.00	5.00
Fish meal	1.40	1.40	1.40	1.40
Vitamin & mineral premix <sup>1</sup>	2.33	2.33	2.33	2.33
Fish oil <sup>2</sup>	9.00	9.00	9.00	9.00
<b>Immunomodulator</b>				
Levucell		0.50		
<i>Schizochytrium</i>			1.00	
VST				0.10
<b>Proximate analysis</b>				
Protein (%)	50.6	51.2	49.3	49.4
Lipid (%)	15.3	15.3	15.7	15.4
Ash (%)	10.0	9.6	9.7	9.8
Moisture (%)	7.2	7.3	7.1	7.2

<sup>1</sup> The vitamin and mineral premixes supply the following concentrations (mg/Kg diet): choline chloride, 3500; ascorbic acid, 2000; biotin, 0.6; B<sub>12</sub>, 0.6; folic acid, 16.5; inositol, 132; vitamin K, 9.2; niacin, 222; pantothenic acid, 106; pyridoxine, 30.9; riboflavin, 52.9; thiamin, 43.0; vitamin E, 503 IU; vitamin A, 6614 IU; vitamin D, 441 IU; zinc, 75; manganese, 20; copper, 1.5; and iodine, 10.

<sup>2</sup> Fish oil stabilized with 500 mg of liquid ethoxyquin per kg of oil; 4% oil added during pelleting and the remainder as a top dressing.

Table 4. Dosage of the immunomodulators.

Compound	Dosage (% of diet)	Dosage (mg/kg fish)
Control	Modified Abernathy Diet	— — —
VST	0.1% “ ”	20 mg/kg fish
<i>Schizochytrium</i>	1.0% “ ”	200 mg/kg fish
Levucell	0.5% “ ”	100 mg/kg fish

Table 5. Proximate composition of the fish in the lipid level study

Diet	Protein	Lipid	Moisture	Ash
Low fat (9%)	16.2 (69.5*)	4.7 <sup>a</sup> (20.0)	76.7 <sup>a</sup>	1.9 (8.2)
Medium fat (11.4%)	16.1 (70)	5.2 <sup>a</sup> (22.6)	77 <sup>a</sup>	1.9 (8.3)
High fat (19.9%)	16.3 (64.7)	7.4 <sup>c</sup> (29.4)	74.8 <sup>b</sup>	2.0 (7.9)
Control (15.7%)	16.3 (64.2)	6.7 <sup>b</sup> (26.4)	74.6 <sup>b</sup>	2.1 (8.3)

Values are means of 4 replicates, five fish were pooled from each tank. Means within the same column with different superscripts are significantly different ( $P < 0.05$ ).

\* Numbers in parentheses are on a dry matter basis

Table 6. Liver glycogen, triglycerides and hepatosomatic index (HSI) of the fish in the lipid level study

Diets	Glycogen ( $\mu\text{g}/\text{mg}$ wet wt)	Triglycerides ( $\mu\text{g}/\text{mg}$ wet wt)	HSI
Low fat (9%)	3.64	2.51	1.20
Medium fat (11.4%)	5.48	2.78	1.17
High fat (19.9%)	4.07	2.19	1.18
Control (15.7%)	4.76	2.19	1.07

Values are means from 4 replicates, five fish from each tank were analyzed.

Table 7. Liver glycogen, triglycerides and hepatosomatic indexes (HSI) of the fish fed the semi-moist diets



# Development of Open Formula Diets and New Feeding Strategies

Diets	Glycogen ( $\mu\text{g}/\text{mg}$ wet wt)	Triglycerides ( $\mu\text{g}/\text{mg}$ wet wt)	HSI
Semi #1	8.81 <sup>c</sup>	4.05	1.07
Semi #2	18.39 <sup>b</sup>	6.53	1.20
Semi #3	52.53 <sup>a</sup>	5.48	1.31
Control	6.80 <sup>c</sup>	5.93	1.04

Values are means from 4 replicates, five fish from each tank were analyzed.

Means within the same column with different superscripts are significantly different ( $P < 0.05$ ).

Table 8. Percent weight gain of the fish in both glucan studies. The weight gain was determined for the 14 day dose period

Treatment	1st study % weight gain	2nd study % weight gain
Control	25.4	6.2
Levucell	30.2	23.9
<i>Schizochytrium</i>	32.3	19.0
VST	38.8	17.1

Table 9. Final skin, nare and vent lysozyme levels taken at 50 % mortality (1st study) or at the termination of the second study

Treatment	Skin		Nare		Vent	
	1st	2nd	1st	2nd	1st	2nd
Control	15.3	38.3	304.2	260.6	167.6	172.7
Levucell	15.4	50.0	252.3	275.0	153.1	205.8
<i>Schizochytrium</i>	9.5	43.0	190.2	235.7	163.1	192.3
VST	14.2	41.0	276.0	250.1	163.6	203.5

Gannam

## Evaluation of the Influence of Diet and Demand Feeding on Fish Performance and Phosphorus Discharge<sup>1</sup>

Ronney E. Arndt, Eric J. Wagner<sup>2</sup>, Charles R. Bobo, Patrick A. Brown,  
Ronald L. Roubidoux, M. Douglas Routledge, and Quentin A. Bradwisch

*Fisheries Experiment Station, Logan, Utah 84321*

Three separate studies were conducted to determine the influence of diet type (floating feed, low phosphorus feed, standard sinking feed) and feeding method (hand vs demand) on the hatchery performance of rainbow Oncorhynchus mykiss and cutthroat trout O. clarki utah. In the first study, a production-scale evaluation of a low phosphorus fish feed was conducted at three of Utah's state hatcheries. At the Mantua, Loa, and Midway hatcheries, rainbow trout were fed either a standard grower diet (control) or a low phosphorus (low-P) diet. Fish fed the low-P diet grew better than the control group at the Loa and Midway hatcheries, but at Mantua the opposite was true. Total weight gain, specific growth rates, and feed conversion ratios were better for the low-P groups at Loa and Midway, while at Mantua weight gain and feed conversions were better for the control group. At all three hatcheries there was some variability among indices measured by the Health Condition Profile, but no trends suggesting that low phosphorus diets compromised fish health. Fish fed the low-P diet at Midway, which has hard water, revealed a reduction in lithic deposits of the kidney and psuedobranch. Total P discharges measured at the raceway tails showed a reduction of 38% at Mantua, 27% at Loa, and 25% at Midway with fish fed the low-P diet compared to the control diet. The average feed cost was \$US 0.76/kg fish for low-P feed and \$ 0.74/kg fish for the control diet.

In the second study, rainbow trout were fed one of four commercial (Silvercup) diets: floating trout (TF), steelhead (SF), sinking low-P (LP), or salmon (S) formulations. Fish fed the SF diet had consistently better final weights, total weight gain, specific growth rates, and feed conversion ratios compared to the LP or TF treatments. Feed cost per kg fish produced was not influenced by diet type and averaged US\$ 0.61. Diet type did not influence the health of the fish nor did it significantly influence the degree of fin

erosion exhibited by the fish. Midway through the study, total phosphorus discharge was considerably higher for the SF and LP fish, 65 and 60 mg P/day/kg fish respectively, compared to 36 and 31 mg P/day/kg fish for the TF and S treatments respectively. By the end of the study, the LP fish had higher total phosphorus in raceway effluent, 76 mg P/day/kg fish, compared to 44 and 42 mg P/day/kg fish for the TF and SF fish respectively.

In the third study there were two separate feeding trials, one for cutthroat trout, and the other for rainbow trout. Each specie was fed an extruded floating feed either by hand or a demand feeder, or a sinking pelletized feed. For both feeding trials, measurements of the total phosphorus in raceway effluent revealed a trend of increasing phosphorus concentration ranked by treatments as follows: floating feed by hand < floating feed by demand feeder < sinking feed by hand. Final mean weights of cutthroat trout were not significantly different. However, fish fed the floating feed via a demand feeder had significantly better feed conversions, 0.69, than the fish fed the floating feed by hand, 0.85, or the sinking feed by hand, 0.83. Rainbow trout fed the floating feed via a demand feeder had significantly larger final weights, 74.4 g/fish, compared to 63.8 g/fish for the floating feed by hand treatment, or 58.8 g/fish for the sinking feed by hand treatment. Feed costs per kg fish produced were not significantly different between treatments within a given trial and averaged US \$0.83 for the cutthroat and \$0.76 for the rainbow.

It appears that the use of either low-P or floating feeds fed by hand may be a good way to reduce phosphorus discharges from hatcheries while still maintaining fish production at a reasonable cost.

---

<sup>1</sup> This paper was submitted too late to be included in the program but the abstract is being included in the Proceedings at the request of the authors.

<sup>2</sup> Paper would have been presented by Mr. Wagner if time would have permitted.



# Session II

## **External Influences**

Session Chair:

Herb Pollard

(National Marine Fisheries Service)

# A Trout's Perspective: Migration Patterns of Volitionally Released Hatchery Trout in the Elochoman River

Charmane Ashbrook & Howard Fuss

*Washington Department of Fish and Wildlife, Hatcheries Program,  
600 Capital Way N., Olympia, WA 98501-1091*

## Introduction

While we cannot ask a trout how it perceives migration, through observation we can determine if fish migrating from a rearing pond exhibit behavior(s) that indicate whether they will migrate to the ocean or remain in freshwater. Hatchery fish which remain in the river (residualize) are potential predators or competitors with wild fish. As mandated by the Endangered Species Act, hatchery operations must meet certain limitations for adverse impacts on wild populations. Therefore, it is important to learn about the post-release behavior of hatchery reared fish 1) to understand if this behavior is affected by how the fish are released from the hatchery, and 2) to know if we can determine prior to release whether some fish are apt to become residuals.

Schuck et. al. (1993) have studied summer steelhead migrating from a rearing pond on the Tucannon River in eastern Washington. Their work indicates that there are at least two populations of summer steelhead within the rearing pond: one group that actively migrates and one group that prefers to remain in the rearing pond. The latter fish are more apt to residualize in the river if forced from the rearing pond. To learn if a similar situation exists in the western Washington populations, we have been studying the migratory behavior of steelhead and cutthroat trout released from Beaver Creek Hatchery. This hatchery is located on Beaver Creek, about 0.5 miles from the confluence with the Elochoman River, which is near the town of Cathlamet, Washington.

Beaver Creek Hatchery releases approximately 150,000 winter steelhead, 30,000 summer steelhead, and 30,000 sea-run cutthroat trout from a gravel rearing pond (dimensions: 750 feet x 100 feet). Prior to our study, the typical manner of release was to pull a few dam boards beginning in April and continue removing a few more each day for about one week. Although this release was considered volitional, the pond was in fact lowered over a one week period and the fish were mostly flushed out. The remaining fish were forced out by hatchery workers. This method of release was revised in 1995 to accommodate our study design so that the fish were allowed to migrate from the pond over a longer period of

time. In 1996 and 1997, we allowed the pond to remain near full level to create a more realistic volitional release. The fish were allowed to migrate from late April through May. We were seeking answers to the following questions:

1. Can we influence post-release residualism through a volitional release?
2. Do the residuals come from one portion of the release or are they homogenous throughout the release?
3. Does release timing affect cutthroat survival to adulthood?

By marking initial and late migrants of a volitional release, we could obtain answers to these questions through snorkel surveys and observation of adults that returned to the hatchery to spawn. In this paper we discuss the results from cutthroat trout.

## Methods

An approximately 100 foot long pipe connects the rearing pond to a raceway. The raceway is connected to Beaver Creek by an approximately 30 foot long pipe. All steelhead and cutthroat trout leaving the rearing pond are counted by a conductivity electronic fish counter (Smith-Root, Inc.; model SR-1600) which is located in the raceway at the end closest to the rearing pond. This set-up enables us to capture fish that have volitionally left the rearing pond in the raceway. Table 1 shows the first and last day of release for migration years 1995-1997.

**Table 1:** First and final day of release of cutthroat trout from Beaver Creek rearing pond by migration year.

Migration Year	First Day of Volitional Release	Last Day of Volitional Release
1995	April 25	May 3
1996	April 24	May 16
1997	April 29	May 21

In 1995, we collected weight and length data on fish that migrated on the first day (April 25) and last day (May 3) of the release. Using a needle with dental floss as thread, we applied a dental floss tag by sewing the dental floss through the posterior portion of the dorsal fin and tying it off. Floss tags were applied to a portion of the initial migrants. Different colors of floss were used to distinguish steelhead and cutthroat. An anesthetic, tricaine methanesulfonate or MS-222, was given to the trout via the water they were held in prior to tagging. After tagging, trout recovered in buckets of fresh water prior to being released. Snorkel surveys occurred one to three times per week after release until October, 1995, to observe whether trout residualized in the Elochoman River. We did not apply floss tags to any of the late migrants because we learned that the tags frayed and came off of the fish easily, making them unsuitable for differentiating fish while doing snorkel surveys.

In 1996, we collected weight and length data on the first day of release (April 24) and on the last day of release (May 16). We used a new tagging method, the visual implant jet tag developed by Dan Thompson of Washington Department of Fish and Wildlife (WDFW) to differentially tag the initial and late migrants. The jet tag material, a pigmented polymer, is injected into the anal fin with a needleless injector under high pressure and results in a visible mark that can be seen with the naked eye. This met our requirements for a different colored mark that was visible while snorkeling and could be applied to each migrating group. Further, this tag was relatively easy and inexpensive (\$0.04 per tag) to apply. A Quonset hut was set up next to the raceway with tagging stations. Fish were collected out of the raceway by net and held in buckets. An anesthetic, tricaine methanesulfonate (MS-222), was given to the trout via the water they were held in prior to tagging. After tagging, trout were released back into the raceway for recovery. Our method of collecting and holding fish during the initial tagging may have been stressful to the fish, which may have affected results. Red jet tags were applied to a portion of initial migrants (April 24-29) and yellow jet tags were applied to a portion of late migrants (May 15-16). Tagging of the late migrants occurred with less stress to the fish because we collected fewer fish at a time and changed the water they were held in more often prior to tagging them.

Most of the fish in the late migrant group were actively migrating from the pond. Snorkel surveys occurred two times per week until October, 1996, to observe whether

trout residualized in the Elochoman River. Hook and line sampling also occurred to locate residuals. Adult cutthroat that returned to spawn in 1996 were examined for jet tags to obtain a survival estimate.

In 1997, we collected weight and length data on the first (April 29) and last (May 21) day of release. We again applied red jet tags to a portion of initial migrants (April 29-30) and yellow jet tags to a portion of late migrants (May 20-21). In 1997, a mass marking trailer was used to tag the trout so as to reduce stress to the fish and increase efficiency. The late migrants consisted of fish that were either migrating or residing in the pond. Snorkel surveys occurred two times per week until October, 1997. Hook and line sampling also occurred to locate residuals. As in 1996, we examined adult fish returning in 1997 to obtain a survival estimate.

## Results

### Migration

Table 2 shows the number of fish released from the rearing pond during the months of April and May. Because cutthroat and steelhead were mixed together in the pond, these numbers are estimates. To obtain the number of cutthroat that migrated in April, we calculated the estimate by multiplying the ratio of cutthroat to steelhead migrants that were tagged by the total amount of steelhead and cutthroat that migrated during the month of April. We subtracted this number from the total number of cutthroat migrants to obtain the number of cutthroat that migrated in May.

**Table 2:** Estimated number of cutthroat migrants by month and year from Beaver Creek rearing pond. Number in parentheses indicate the number of days each month the fish were allowed to migrate.

Migration Year	April	May
1995	35,591 (6)	5,326 (3)
1996	6,066 (7)	18,201 (16)
1997	5,649 (7)	25,607 (21)

### Size of Migrants

Table 3 shows the lengths and condition factors of cutthroat trout for migration years 1995-1997. Fish in the

## Migration Patterns of Volitionally Release Hatchery Trout

initial portion of the 1995 migration were significantly longer ( $P < 0.05$ ) than fish in the middle or final portion of the migration. Similar to 1995, 1996 initial migrants (April 23) were significantly longer ( $P < 0.05$ ). A portion of 1997 initial migrants was weighed and measured on April 29<sup>th</sup> and final migrants on May 21<sup>st</sup>. Table 3 shows the lengths of the initial and final migrants. As in the previous two years, the initial migrants were significantly longer than the final migrants ( $P < 0.05$ ).

To examine the smoltification of the trout, we also looked at condition factor (Table 3). We found no difference in condition factor among the 1995 initial and final migrants, although the fish in the final portion were slimmer than fish in the other two groups. Cutthroat trout initial migrants in 1996 were significantly more robust than final migrants (May 16)( $P < 0.05$ )(Table 3). For 1997, there was no difference in initial and final migrant condition factors among the groups (Table 3).

### Snorkel Surveys and Hook and Line Sampling Results

- ▶ We did not see any trout in the Elochoman River with dental floss tags in 1995.
- ▶ In our snorkel surveys in 1996, we did not see any yellow tagged (late migrant) cutthroat trout in the river.
- ▶ During snorkel surveys in 1997, we saw only yellow tagged (late migrant) cutthroat trout in the river. The greatest number of jet tagged cutthroat trout we saw in one day was 3 fish. Because 697 cutthroat trout were given a yellow tag, the rate of residualism (#observed/ # tagged) for yellow jet tagged cutthroat trout in 1997 was 0.43%.
- ▶ By the end of September in both 1996 and 1997, we no longer observed jet tagged trout in the Elochoman River.
- ▶ In 1996, we caught one red jet tagged cutthroat (325 mm). Because 454 cutthroat trout were given a red jet tag, the rate of residualism (#observed/ # tagged) for red jet tagged cutthroat trout in 1996 was 0.22%.
- ▶ In 1997, we did not obtain any jet tagged cutthroat through hook and line sampling.

### Adult Returns

Only yellow jet tagged cutthroat trout returned to the hatchery in 1996. The survival rate to escapement was 1%. Table 4 shows the length of the returning cutthroat adults in 1996. Of the adult return in 1997, 18 were initial migrants and 10 were final migrants. Table 5 shows the length of the returning cutthroat adults in 1997.

**Table 3:** Length data and condition factor of cutthroat trout released from Beaver Creek Hatchery in 1995, 1996, and 1997.

Group	Migration Year	Initial	Final
Length (mm)	1995	219	204
	1996	222	202
	1997	226	210
STD	1995	26.5	24.6
	1996	18.8	22.1
	1997	19.1	25.7
CV (%)	1995	12.1	12.1
	1996	8.44	10.9
	1997	8.47	12.3
Condition Factor	1995	0.828	0.816
	1996	0.892	0.811
	1997	0.889	0.872
STD	1995	0.100	0.0799
	1996	0.100	0.100
	1997	0.281	0.0935
CV (%)	1995	12.1	9.80
	1996	10.2	13.0
	1997	31.8	10.7

**Table 4:** Length at return of initial, late, and untagged cutthroat to Beaver Creek Hatchery in 1996.

Group	Late Migrants	Untagged
Length (mm)	351	348
STD	39.1	27.2
CV (%)	11.1	7.8



**Table 5:** Length at return of initial, late, and untagged cutthroat to Beaver Creek Hatchery in 1997.

Group	Initial	Late	Untagged
Length	344	318	334
STD	17.9	27.1	33.7
CV (%)	5.20	8.51	10.1

Because 1,872 initial cutthroat migrants were given a red jet tag compared to 695 final cutthroat migrants, the return rate was 0.962% for initial migrants and 1.44% for final migrants. A chi square test indicated these results were not significant ( $p < 0.05$ ). Initial migrants that measured greater than 220 mm consisted of 63.6% of the sample while final migrants greater than 220 mm were 33.3% of the sample. There were significant differences in mean length of returning adults. Fish from the initial migrants were longer than either the final or untagged returns, and untagged fish were significantly longer than final migrants ( $p < 0.05$ ).

#### Discussion:

Prior to 1996, trout were released "volitionally" over a one week period from Beaver Creek rearing pond. Historically, the majority of trout released from Beaver Creek Hatchery occurred in April. As of 1996, the volitional releases from this pond occurred over at least 30 days to allow the trout a greater time period to migrate. As of 1996 and 1997, the majority of the fish left in May, with the 50% migration occurring on May 12 and May 13, respectively. In both years, by the end of May, there were some fish still in the pond that did not migrate. Some fish from this tail end of the migration period residualized in the river. In 1997, we saw 3 yellow jet tagged trout, as compared to 1 in 1996. In 1997, we marked the final migrants one week later than in 1996, and we noticed that some of the fish in this group resisted moving into Beaver Creek. Because of this, we believe a portion of this group were not ready to migrate relative to the group of final migrants marked in 1996.

The red and yellow tags are visible underwater and remain on the cutthroat well enough to identify them when they return to the hatchery as adults. Studies are currently being done by WDFW to learn if the presence of jet tags affect survival of fish to adulthood (Dan Thompson, WDFW, personal communication.)

The fish that migrated last or were forced from the pond were the only tagged fish seen in snorkel surveys and the only fish that returned to spawn in 1996. Also, the final migrants in 1997 survived better than the initial migrants. This indicates cutthroat at Beaver Creek do not survive as well when released in late April compared to May.

Schuck et. al.'s (1997) application of passive integrated transponder (PIT) tags revealed that the most successful smolts were the largest, leanest fish. Tipping (1995) found that condition factor is more reliable at predicting survival to adulthood than size. Our results mirror the results of Tipping's study because the fish with the best survival were the shortest and the leanest. However, fish released in Tipping's (1995) study were released as a single group, therefore date of release was not considered as a variable.

Another interesting explanation of why the final migrants survived better can be found by looking at the bird populations in the lower portion and estuary of the Elochoman River. Roby (1997) has found that Caspian Terns and Cormorant populations are increasing in size on the lower Columbia River and that these birds prefer yearling sized smolts. We noticed that as the initial migrants were being released, the piscivorous birds began to increase as well at the mouth of the river, particularly at low tide. The final migrants in both 1996 and 1997 were released as the bird populations were declining (Fuss et. al., 1996, 1997).

Shuck et. al. (1997) stated, "managing acclimation ponds to retain potentially residual juveniles reduces the presence of these fish in the river and their potential impact on wild salmonids." If we can distinguish migrants from non-migrants, management practices could change to reduce post-release residualism and use non-migrants in put and take trout fisheries in lakes without access to anadromous waters.

Next year we plan to jet tag initial and final migrants again. We will separate the final migrants into two groups: an actively migrating and a non-migrating group. Each group will be jet tagged with a unique color to allow us to identify these fish should they residualize and to calculate separate survival estimates.

#### Conclusions

1. Initial cutthroat migrants are bigger and have higher condition factors than final migrants.

## Migration Patterns of Volitionally Release Hatchery Trout

2. Final cutthroat migrants are more likely to residualize in the Elochoman River.
3. Final cutthroat migrants are more successful at returning to Beaver Creek Hatchery.

### Acknowledgments:

We would like to thank the National Marine Fisheries Service (NMFS) for their funding of this project, the Beaver Creek Hatchery crew for their support and assistance with every step of this project, and Dan Thompson, Dinette Aho, Lynn Andersen, Jim Byrne, Susie Jackson, Eric Larson, Karen Kloempken, Dave Knutzen, and Geraldine VanderHaegen for assistance with jet tagging and examining returning adults.

### References

- Fuss, H., C.E. Ashbrook, and J. Byrne. 1996. Mitchell Act Hatcheries Evaluation: Annual Report, Part I. Report No. H96-04. Washington Department of Fish and Wildlife.
- Fuss, H., C.E. Ashbrook, and J. Byrne. 1997. Mitchell Act Hatcheries Evaluation: Annual Report, Part I. Report No. H97-04. Washington Department of Fish and Wildlife.
- Martin, S. W., A.E. Viola and M. L. Schuck. 1993. Lyons Ferry Evaluation Study: Annual Report 1993. Washington Department of Fish and Wildlife Report to the U.S.F.W.S. Report No. AFF 1/LSR-93-1.
- Roby, Dan. 1997. Personal communication, 19<sup>th</sup> Annual Smolt Workshop, Oregon State University.
- Schuck, M.L., A.E. Viola and J. Dedloff. 1997. Lyons Ferry Evaluation Study: Annual Report 1997. Washington Department of Fish and Wildlife Report to the U.S.F.W.S. Report No. H97-08.
- Tipping, J., R. Cooper, J. Byrne, and T. Johnson. 1995. Length and condition factor of migrating and non-migrating hatchery reared winter steelhead smolts. *Progressive Fish Culturist*; 57: 120-123.



# Pinniped Scarring at Beaver Creek, a Lower Columbia River Hatchery

Jim Byrne

Assessment and Development Division, Hatcheries Program,  
Washington Department of Fish and Wildlife, 600 Capitol Way N., Olympia, WA 98501

**ABSTRACT** — Adult returns of winter steelhead *Oncorhynchus mykiss* and sea-run cutthroat *O. clarki* have been monitored at Beaver Creek Hatchery since 1983. Returning adults are examined for seal and net marks, when they are removed from the hatchery trap, prior to being placed into adult holding ponds by hatchery personnel. Scars are left primarily by the Pacific harbor seal, *Phoca vitulina* and California sea lion, *Zalophus californianus*. Numbers of these predators have been increasing since the early 1940's. Steelhead showed distinct patterns of pinniped scarring. The preferred target site is the lower ventral area, containing the viscera and gonads. Over the 14-year time frame, winter steelhead had a significant correlation ( $r=0.55$ ,  $r^2=0.31$ ,  $p=0.040$ ) toward increased scarring from pinnipeds. Percent scarring and winter steelhead brood year survivals were negatively correlated ( $r=-0.70$ ,  $r^2=0.49$ ,  $p=0.025$ ). Sea-run cutthroat, while showing similar scar patterns, had a significant correlation ( $r=-0.56$ ,  $r^2=0.31$ ,  $p=0.039$ ) to a reduced incidence of scarring. The difference in scar rate may be due to the timing of fish entry to the Columbia and Elochoman Rivers. Recent cutthroat median trap dates (mid-November), indicate cutthroat migrate prior to the highest concentrations of harbor seals and sea lions (mid-December through mid-March). Winter steelhead median trap dates occur from late-December through mid January, placing them within the greatest concentrations of pinnipeds.

Pinnipeds common in the lower Columbia River are primarily Pacific harbor seals *Phoca vitulina* and California sea lions *Zalophus californianus*. Due to the security provided by the Marine Mammal Protection Act, their numbers have been increasing in recent years. Large congregations have been reported in the Columbia River Estuary and they have been seen up the Columbia River as far as Willamette Falls 128 miles and Bonneville Dam 145 miles upriver. To a lesser extent, populations of the threatened Stellar sea lions *Eumetopias jubatus* also are present.

The Marine Mammal Protection Act was passed in 1972. The California sea lion population has been increasing at an annual rate of about 5% per year and harbor seals 5-7% since the mid-1970s. Sea lions breed and pup primarily on California's Channel Islands, with male sea lions migrating northward into Washington and Oregon each year from September to May. The most famous probably are Hondo and Hershel at the Ballard Locks ship channel in Seattle. The 1994 west coast sea lion estimate was 161,000. This may be greater than historical levels (Low, 1991). In the Columbia River, sea lions (300-500) occur in fall, winter and spring. Sea lions are opportunistic feeders, with a diverse diet, and will move into specific areas in response to local prey abundance (NMFS, 1997).

The 1993 Washington estimate for Harbor seals was 34,134 and 9,251 for Oregon. Seals in the Columbia are present year round, but peak numbers (>3,000) occur mid-December through mid-March. Many of the seals, which pup in the coastal estuaries of Washington and Oregon in summer, migrate to feed in the Columbia River in spring and fall, when salmonids are present. Highest

seal counts occur in winter, when eulachon (smelt) are abundant (NMFS, 1997). Seals are opportunistic feeders, relying on items of local and seasonal abundance.

In past years, eulachon has been the main component of Columbia River sea lion diet, as seen in scat and intestinal tract sampling. As eulachon numbers have declined in recent years, pinnipeds have diverted their attention to other prey species. In 1985, Beach et al. (1985) reported salmonids in 13% of sea lion gut samples. A decade later, Brown et al. (1995) reported salmonids in 28% of samples investigated. Data is not as clear for harbor seals, due to different sampling periods over the years. In winters of 1986-88, salmonids were not seen in gut samples (Brown et al., 1995). But in September through October of 1994, salmonids were noted in 39% of scat samples investigated (Reimer and Brown 1996).

NMFS in 1977 estimated a minimum total biomass consumed by sea lions and seals in Washington, Oregon and California of 217, 400 metric tons. This is approximately half of the commercial fisheries harvest in these states. This includes all biomass, not just salmonids. For the Columbia River, sea lion consumption is estimated at 390 tons. Estimated harbor seal consumption for the Columbia River, Tillamook Bay and the coast north of Yaquina Bay is 3,851 tons. There is little information on pinniped interactions on smolts moving seaward, because of their smaller size and submerged ingestion. Salmon and seal interactions in the open ocean are believed to be minimal (Fiscus, 1979 and 1980). At the Ballard Locks in Seattle, it was estimated that 65% of the winter steelhead run was consumed by sea lions (Scordino and Pfeifer, 1993).

Although this paper focuses on pinniped interactions with adult salmonids, there are also concerns over juveniles. Pinnipeds are still present, in the Columbia system, after the eulachon run ends; during the spring smolt migration. Fall chinook smolts may be too small to acquire pinniped interest, but steelhead, cutthroat and yearling spring chinook are in the size range of pinniped prey.

Beaver Creek Hatchery is located on the Elochoman River approximately 30 miles from the Columbia's

mouth. Staff biologists have viewed seals at the mouth of the Elochoman and in the Elochoman slough. Seal scarring has been reported at a variety of other facilities (Table 1.)

Beaver Creek has maintained records of pinniped scarring on returning fish (Table 2) since 1983. Upon capture in the hatchery trap, fish were examined daily by hatchery staff for species, sex, net and seal marks prior to being placed in adult holding areas. A graphical representation is presented in Figure 1.

Table 1. Pinniped caused scarring observed in Oregon winter steelhead hatcheries (NMFS, 1997).

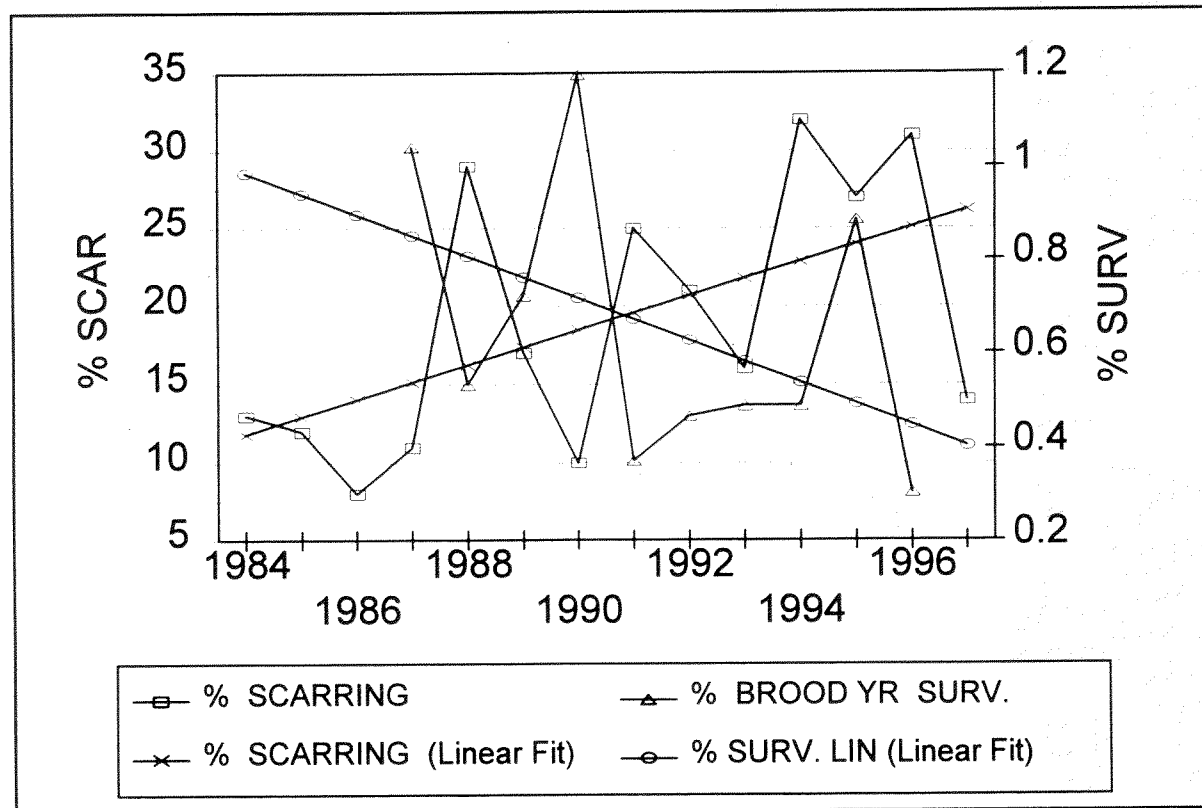
Location	Time frame	Incidence of Scarring
Nehalem River Hatchery	1985-1992	6.4-53%
Alsea and Fall Creek Hatcheries (Alsea River)	1989-1992	19-27%
Salmon River Hatchery	1984-1986	38-40%
Big Creek Hatchery (lower Columbia River)	1992-1993	22-43%
Klaskanine Hatchery (lower Columbia River)	1992-1993	20-52%
Cedar Creek Hatchery	1989-1992	10-43%
Trask River Hatchery (Tillamook Bay)	1989-90	35%
Bandon Hatchery	winter 1990-91	26%
Elk River Hatchery	winter 1990-91	21%
Marion Forks Hatchery (Willamette River)	winter 1990-91	10%
South Santiam Hatchery (Willamette River)	winter 1990-91	12%
Rock Creek Hatchery	winter 1990-91	45%

There was a statistically significant ( $r=0.55$ ,  $r^2=0.31$ ,  $p=0.040$ ) correlation of increasing seal (refers to all pinnipeds) scarring over time at this facility. Years of reduced scarring, (<15%) were also years of highest returns (>1000 fish). With larger numbers of fish available, similar levels of attack would result in a smaller proportion of scarred fish.

Beaver Creek has a low percentage of three salt adults 2-11%. If percent of scarring is contrasted with the brood year percent return for 1986-87 through 1995-96 a

significant negative ( $r=-0.70$ ,  $r^2=0.49$ ,  $p=0.025$ ) linear regression exists. This is shown graphically in Figure 1. Seal marks have been consistently recorded at Skamania Hatchery, another WDFW facility rearing summer steelhead since 1992. When marks were recorded, the incidence was much less than for winter steelhead at Beaver Creek. Summer steelhead migrate prior to pinniped concentration in the Columbia. In addition, seal wounds are observed at sorting time (late-November) prior to spawning; not at capture, and some wounds might have healed in the interim.

## Pinniped Scarring at Beaver Creek, A Lower Columbia River Hatchery



**Figure 1.** Beaver Creek winter steelhead percent seal scarring and brood year percent survival 1984-1996.

Beaver Creek data were not analyzed by sex. Since 1992, Skamania seal marks were recorded by sex and are presented in Table 3. For all years, females showed a greater incidence of scarring than males. At Beaver Creek, the impression is that females also have a much greater incidence of scarring than males. This may indicate that pinnipeds have a preference for gravid females at this time of year. Beaver Creek will begin to sex scarred fish during the 1997-98 spawning season.

At both facilities, the greatest incidence of pinniped marks occurred in the mid-ventral area. This was also the most frequently targeted area reported for gillnetted chinook (Beach et al., 1985)

### Sea-run Cutthroat

Records of pinniped scarring on sea-run cutthroat are presented in Table 4. There was a significant ( $r=-0.56$ ,  $r^2=0.31$ ,  $p=0.039$ ) trend toward decreased pinniped

scarring over time. Unlike winter steelhead, which showed increased scarring over time, cutthroat had decreased incidence of seal wounds. This can be seen graphically in Figure 2.

Additionally, a linear regression was run on the percent scarred versus total yearly return. Total return was examined, since cutthroat have a complicated life history with multiple spawnings. A non-significant regression existed ( $r=0.43$ ,  $r^2=0.18$ ,  $p=0.129$ , Figure 2).

Although harbor seals were present year-round in the Columbia River, peak concentrations ( $>3,000$ ) occur from mid-December through mid-March. Three to five hundred sea lions are also present in fall through spring (NMFS, 1997). Beaver Creek winter steelhead median trapping dates ranged from December 27 through January 24, and steelhead showed increased scarring over the fourteen year time frame. The median trapping date is the date that 50% of the total run arrives at the hatchery.

Table 2. Percentage of returning Beaver Creek winter steelhead bearing evidence of pinniped scarring.

Year	Hatchery Return	Number Scarred	Percent of Run
1983-84	2,126	270	13
1984-86	1,802	224	12
1985-86	1,106	87	8
1986-87	1,233	141	11
1987-88	553	158	29
1988-89	771	128	17
1989-90	1,235	127	10
1990-91	393	99	25
1991-92	426	91	21
1992-93	522	81	16
1993-94	546	174	32
1994-95	800	217	27
1995-96	405	127	31
1996-97	476	65	14

Table 3. Incidence of Skamania summer steelhead pinniped scarring by sex 1992-97.

Year	Male	Female
1992	1.0%	3.0%
1993	0.0%	2.0%
1994	0.9%	1.0%
1995	4.0%	10.0%
1996	6.0%	7.0%
1997	12.0%	17.0%

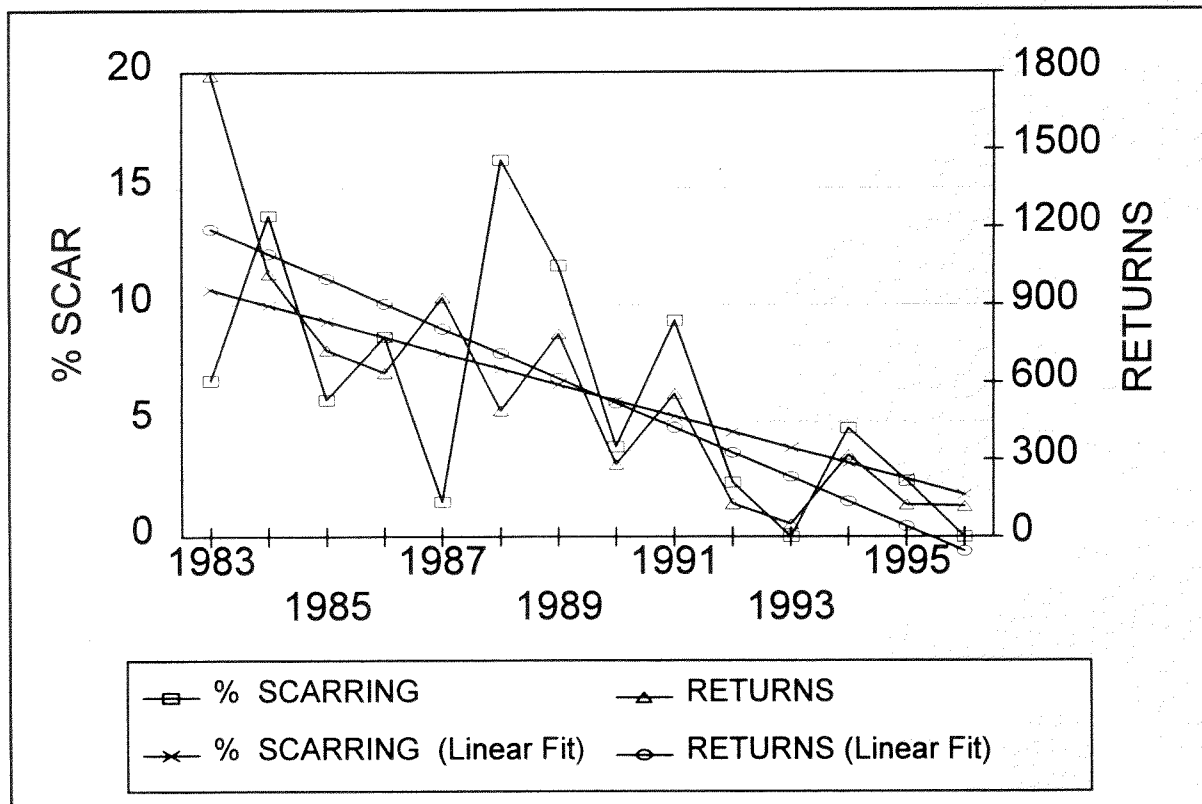
Table 4. Percentage of returning Beaver Creek sea-run cutthroat bearing evidence of seal scarring.

Year	Hatchery Return	Number Scarred	% Scarred
1983-84	1789	121	7
1984-85	1020	141	14
1985-86	726	43	6
1986-87	637	55	9
1987-88	929	14	2
1988-89	493	80	16
1989-90	788	92	12
1990-91	282	11	5
1991-92	558	52	9
1992-93	128	3	2
1993-94	49	0	0
1994-95	319	15	5
1995-96	124	3	2
1996-97	118	0	0

This is not the case for Beaver Creek sea-run cutthroat, which had a significant correlation towards reduced scarring. This could be due to the difference in run timing between winter steelhead and sea-run cutthroat. Cutthroat appear to pass through the Columbia system and into the Elochoman River before the main body of pinnipeds arrive. Median cutthroat trap dates for this period ranged from November 12 through January 10. The last two years had low incidence of scarring, coinciding with the earliest median trap dates (November 12 & 14). Over the 14 year time frame, there was no correlation ( $p=0.99$ ) between cutthroat median trap date and percent scarring.

Cutthroat enter the Elochoman approximately two months earlier than winter steelhead, prior to large concentrations

## Pinniped Scarring at Beaver Creek, A Lower Columbia River Hatchery



**Figure 2.** Beaver Creek sea-run cutthroat percent seal scarring and yearly return 1983-1996.

of pinnipeds forming in mid December. Cutthroat with mid-November median trap dates generally have lower incidences of pinniped scarring than fish arriving in late-November or early December.

In conclusion, many factors have been identified as possible causes for the decline of salmonids in the Columbia Basin. They include: dams, logging, development and land use, commercial fishing, hatcheries and irrigation. For Beaver Creek winter steelhead, an overall increasing percentage of returning adults bear evidence of encounters with marine mammals.

We are only able to observe the survivors of these pinniped attacks. We have virtually no data on the number of fish which do not survive these encounters. It would be reasonable to assume if the percentage of survivors is increasing, then, there could be a corresponding increase in the number of victims. Pinniped populations are increasing, while smelt and salmonid populations are experiencing declines. There

are more seals and sea lions to potentially prey upon less fish.

Sea-run cutthroat at Beaver Creek, show an opposite trend toward reduced scarring. Cutthroat arrive prior to the greatest concentrations of pinnipeds forming, and move through the system before pinniped populations reach sufficient size to cause increased incidences of scarring.

I would like to thank the National Marine Fisheries Service for funding Beaver Creek, one of the Mitchell Act Hatcheries and also this study. I would like to thank Stan Woody and the crew of Beaver Creek for their efforts through the years and Steve Jeffries WDFW's marine mammal biologist for his pinniped slides.



### References

- Beach, R. J., A. C. Geiger, S. J. Jeffries, S. D. Treacy, and B. L. Troutman. 1985. Marine mammals and their interactions with fisheries of the Columbia River and adjacent waters, 1980-1982. NMFS-AFSC Processed Rep. 85-04, 316 p. Alaska Fisheries Science Center, Natl. Mar. Fish. Serv., NOAA, 7600 Sand Point Way NE, Seattle, WA, 98115.
- Brown, R. F., S. Riemer, and S. J. Jeffries. 1995. Food of pinnipeds collected during the Columbia River area salmon gillnet observation program, 1991-1994. Oregon Dept. of Fish and Wildlife, Wildlife Diversity Program, Technical Report #95-6-01, 16 p.
- Fiscus, C. 1980. Marine mammal-salmonid interactions: A review. *In* W. McNeil and D. Himsworth (editors), Salmonid ecosystems of the north Pacific, p. 121-132. Oregon State Univ. Press, Corvallis.
- Low, L. 1991. Status of living marine resources off the Pacific coast of the United States as assessed in 1991. U.S. Dep. Commer., NOAA Tech Memo. NMFS-F/NWC-210, 69 p.
- National Marine Fisheries Service (NMFS). 1977. Investigations of Scientific Information on the Impacts of California Sea Lions and Pacific Harbor Seals on Salmonids and on the Coastal Environment of Washington, Oregon, and California. U.S. Dep. Commer., NOAA Tech. Memo. NMFS-NWFSC-28, 172 p.
- Reimer, S. D., and R. F. Brown. 1996. Marine mammal (pinniped) food habits in Oregon. Oregon Dept. of Fish and Wildlife, Wildlife Diversity Program, Technical Report #96-6-01, 26 p.
- Scordino, J., and B. Pfeifer. 1993. Sea lion/steelhead conflict at the Ballard Locks. A history of control efforts to date and a bibliography of technical reports. (Available from Northwest Regional Office, Natl. Mar. Fish. Serv., NOAA, 7600 Sand Point Way NE, Seattle, WA, 98115.

# Movements of Radio-Tagged Cowlitz River Hatchery Winter Steelhead

Jack Tipping <sup>1</sup>

*Washington Department of Fish and Wildlife, Cowlitz Trout Hatchery,  
1181 Spencer Road, Winlock, WA 98596*

In winter 1995-96 and 1996-97, a total of 22 fresh-run adult hatchery winter steelhead arriving at the Cowlitz River Barrier Dam were implanted with radio tags and transported several miles downstream. Movements of the tagged fish was monitored about 3 times per week from a jetboat using Smith-Root receivers and directional and whip antennae. Results showed that of 191 observations, 52% were in closed water areas. Sport harvest accounted for a minimum of 18% of tagged fish with harvest occurring an average of 11 d after tagging. Of remaining fish, most were thought to be dead or had rejected their tag by 20 days post-tagging. A remote data-logger

stationed near the mouth of the Cowlitz in winter 1996-97 indicated that only 7% of tagged fish exited the river; a female at 11 days post-tagging. Results suggest fish managers should examine the need for closed water areas to protect hatchery fish from harvest. Also, hatchery managers might want to examine the time trend between adult fish arrival and spawning; selection of spawning fish soon after arrival could reduce the time fish are available for sport harvest. This might partially explain why the return to the creel of summer steelhead is often greater than winter steelhead.

---

<sup>1</sup> Jack elected to only have the abstract of his talk published in the Proceedings.

## Tipping

# Stream Nutrient Enhancement with Hatchery Diverted Salmonid Carcasses *A Step in Wild Stock Restoration*

Stephen Evans

*Washington Department of Fish and Wildlife, Hatcheries Program,  
600 Capital Way N., Olympia, WA 98501-1091*

## Introduction

Anadromous salmon transport a significant amount of nutrients from the marine environment to Washington's freshwater systems. These nutrients are critical to the ecological processes of the entire watershed. Kline *et al.* (1990) using stable carbon and nitrogen isotopes of marine origin, documented that salmon carcasses provided a significant amount of the nutrients which "fed" stream life. Richey *et al.* (1975) demonstrated that peaks in nutrient levels, carbon fixation, and periphyton biomass occurred following spawning and death of resident sockeye (*kokanee*) in a stream. Koenig and Burkett (1987) suggested that fewer spawners, resulting in reduction in nutrient input, may be partially responsible for the inability of sockeye salmon populations to increase to historic levels in Karluk Lake, Alaska. Bilby and Bisson (1992), showed evidence of increased growth in coho salmon smolts rearing in the presence of salmonid carcasses. Michael (1995) showed the enhancement effects of spawning pink salmon on stream rearing juvenile coho salmon in the Skagit River, Washington. Additionally, research along the west coast of North America and Japan has suggested not only that carcasses support aquatic organisms, but that they benefit the whole watershed ecosystem. Based on these findings, a very productive use of hatchery surplus and spent carcasses from hatchery spawning operations became evident. The use of carcasses to increase stream productivity could directly benefit wild populations of stream rearing anadromous fish, and benefit animals dwelling in associated terrestrial habitat.

## Process

In response to the accumulation of published research since 1991 that points to the large nutrient deficit in many of Washington's watersheds, the Assistant Director of the Hatcheries Program directed staff to 1) scope the feasibility of placing hatchery diverted salmonid carcasses into streams known and suspected to be nutrient deficient, 2) develop a set of guidelines and protocols for statewide application, and 3) develop an information package for State Environmental Policy Act (SEPA)

compliance directed towards program scale operation of an annual nutrient enhancement effort.

Pilot projects were initiated in 10 river basins west of the Cascade Crest. Stream types varied from steep headwater tributaries to mainstem backwater side-channels and associated wetlands. The main objective was to evaluate logistical aspects of various projects and identify costs, carcass distribution potential, legal and easement issues, impacts on hatchery operations, and the potential for volunteer and cooperative support. Secondary to this effort, we would assess the nutrient uptake and ecosystem response in the different stream types and geologic regions.

The pilot projects were all coordinated through WDFW Hatchery Complex Managers with a strong working relationship established with WDFW Volunteer Resource Coordinators, county conservation districts, The Nature Conservancy, regional fisheries enhancement groups, sports clubs, and interested citizens.

Carcass distribution days were coordinated with spawning operations at the source facility with additional staff needs filled by volunteers. Fish were killed, marked, and placed in covered totes for transportation to predetermined sites within the watershed. Upon arrival, the carcasses were placed according to a pre-approved plan in the stream and along stream margins. The carcass distribution plan for each pilot projects was based on a developing set of guidelines and protocols that could be applied across a wide range of stream types and local constraints. They were as follows:

- Treatment streams will be considered that are within the historic anadromous zone of a watershed. Exceptions will be based on research study needs.
- Streams that have historic data sets or ongoing assessment projects, or both, that can be complemented by marine nutrient enhancement will be given a higher priority.

- Streams or stream reaches immediately upstream from municipal water supplies will be considered only with the concurrence of the water purveyor.
- Streams or stream reaches with identified water quality constraints will be avoided ; exceptions will be made only with concurrence of the regulatory entity.
- Streams should have multiple access points to the treatment reaches, (bridges, wet crossings, culvert crossings, etc.) To accommodate appropriate carcass distribution.
- Treatment reaches of streams will be identified with informational signs at established or customary access points. This is to notify the public of the project and explain the presence of fish which are obviously altered by man.
- Carcasses will be used within designated watersheds or Fish Health Management Zones (HMZ) , as identified by WDFW Fish Health Specialists.
- Carcasses will be used from stocks of fish that have been screened for pathogens as prescribed in the "Co-Managers Salmonid Fish Disease Control Policy" of Washington State.
- Carcass distribution will follow a specific plan agreed to among in-basin permitting and/or fish management agencies.
- If tethers or external tags are used, they will be either of a biodegradable material or will be retrieved from the stream following carcass decomposition.
- Transport and deposition of carcasses will be directly supervised by the project leader or their designee and be accompanied by the appropriate documents for transfer.
- Artificial deposition of salmonid carcasses must not create a direct human health hazard.

To develop these guidelines and have them sensitive to the needs of the agency's programs, a technical work group was established. Representatives from the Habitat, Fish Management, Volunteer Services, Wildlife Management, and Hatcheries programs were involved. Additional representatives from the Department of Ecology, Department of Natural Resources, and the Weyerhaeuser Company participated. The guidelines were used to plan the pilot projects carried out in 1996-97. Approximately 17,000 hatchery diverted carcasses were returned to Washington's streams through the 10 pilot projects. As the projects progressed, the guidelines and protocols were modified to accommodate additional needs and constraints.

In February 1997, a one day Nutrient Enhancement Workshop was held in Olympia, Washington to review and update ongoing research in the field of nutrient enhancement being carried out in the Pacific Rim states. Reports were presented on the pilot projects, and discussions on how to improve the process were held. Information gathered at the workshop was incorporated into supporting documents that accompanied a SEPA package that was submitted for public review in April 1997. On May 6, 1997 a "Determination of Non-significance" (DNS) was adopted which enabled us to expand the scope of future carcass distribution projects. A DNS for the nutrient enhancement program meant that following statewide review by local government, Treaty Tribes, and state resource management agencies had concurred that no significant adverse impacts to the environment would occur if the projects were conducted as proposed.

In the 1997-98 spawning season, WDFW has cooperative projects to distribute more than 50,000 carcasses, with an additional 30-40,000 carcasses expected by the end of the season.

For stream nutrient enhancement work to be effective in the long-term, a committed, coordinated effort between local governments, and resource management entities is essential, as placing carcasses in streams will not by itself bring back large runs of anadromous fish.

# The Suquamish Tribe's Approach to Successful Chum Salmon Enhancement

Paul Dorn

*Suquamish Tribal Fisheries Department, PO Box 498, Suquamish, WA 98392*

## Introduction

The Suquamish Tribe initiated a chum salmon enhancement program in 1977 to rebuild salmon populations in east Kitsap County streams. Most of these streams have small fractions of their historic salmon populations, having been heavily impacted by urbanization and other human activities. The larger streams have low flows that range from 5 to 10 cubic feet per second (cfs). The program objective is to restore Tribal chum salmon fisheries on and near the Port Madison Reservation. Cowling Creek Hatchery was constructed to maintain a hatchery run and to support satellite eggboxes installed on selected local streams. Most of the hatchery chum eggs are transferred to these eggboxes. The unfed fry volitionally migrate from the eggboxes, with the adults returning to spawn in their "new" natal streams. The Suquamish Tribe does not direct a terminal fishery on the adult chum returning to Cowling Creek in order to obtain the maximum possible genetic diversity within the hatchery population.

Cowling Creek Hatchery released Hood Canal origin chum in 1977 and 1978, but switched to local Chico Creek stock in 1979 to preserve genetic stock integrity within east Kitsap County. All Hood Canal adult chum returning to the hatchery were destroyed. Chico Creek, located near Bremerton, Washington, was famous for the thirty-nine Orcas that followed, then consumed, most of the chum returning to the stream in 1997. The Chico Creek chum run represents over 90% of wild chum escapement into east Kitsap County.

## Hatchery Design and Management

Cowling Creek Hatchery was designed to be simple to construct and operate. Pre-cast concrete modules were used to build the intake dams on the north and south forks and the south fork rearing pond dam. The intake dams bypass flood water around the settling pond. The fiberglass hatchery incubators are based on the Netarts design and assembled on site by hand. The rearing pond is a natural in-stream earthen pond (Fig. 1). The adult recapture pond was located intertidally with a dam and fish ladder constructed out of sheetpile. The entire

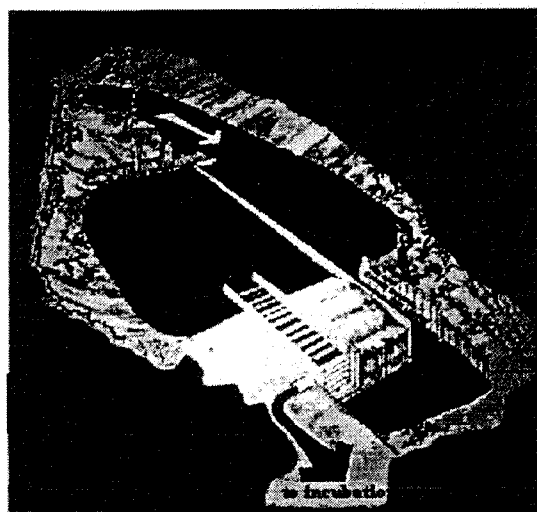


Figure 1. South Cowling schematic.

hatchery was constructed by staff and is designed for gravity flow operation with minimal electrical requirements. The spawning shed is located a short distance above the recapture pond and adult chum are transported to the racks via a custom fish lift. The fish lift is portable and is also used at the Tribe's Grovers Creek Hatchery to move fall chinook.

Adult chum return to the hatchery in late October and continue running into December of each year. The adult chum successfully home in on Cowling Creek's low flow of 100 gallons per minute (gpm). The average winter Cowling stream flow is 400 gpm with the highest flow recorded to date of 20,000 gpm. Limited natural spawning occurs in the intertidal recapture pond because it is saltwater. Hatchery staff seine the pond every weekday and harvest all adults present, up to 1,000 fish/day. Ratios of male to female Cowling chum remain approximately constant between years (Figure 2). Most of the females are ripe and are ready to be fertilized. Excess ripe eggs and any green eggs are sold to the caviar market. Cowling Creek chum are spawned throughout the entire run, with the eggs of two females fertilized by two males in one small bucket. Stream water is introduced to the bucket and the rinsed eggs are transferred to a 5 gallon bucket for water hardening in a 100 parts per million (ppm) iodophor solution for one

hour. The water hardened eggs are transferred to the incubators and remain immersed in ambient temperature surface water. A 1:600 formalin treatment is applied three times a week via a 15 minute drip bottle at the head of each incubator raceway. Fish pathologists inspect 120 adults for viruses and other potential pathogens to certify the stock prior to transferring eggs out of the watershed. Scales are sampled weekly to determine age, and all chum adults are sampled for any tags that may have applied at sea.

Eyed-up eggs are picked, sorted by spawning date, and approximately 2,000,000 are transferred to the satellite eggboxes in proportion to the adult run timing. Approximately 500,000 are hatched in Cowling Creek incubators for release on station (Figure 3).

The early-emerging chum fry are fed in six-foot diameter ponds for several days until they are actively feeding, then released into the 1,000 ft<sup>3</sup> natural pond. All subsequent fry volitionally migrate into the rearing pond without handling except for a few stragglers. The fry initially start feeding at a weight of 0.3 gms and are fed for four to six weeks in an attempt to achieve a 1 gm body weight. Cowling Creek chum fry have not been observed to exhibit a feeding response behavior towards hatchery personnel, but instead randomly school throughout the pond searching for food during their residence in the pond. Several cutthroat trout are usually found in the pond during release, but avian predation is minimized by a birdnet over the pond. Releases occur after midnight on high tide and scuba diving observations verify marine predation is low during the first hours that the fry acclimate to Miller Bay estuary. Approximately one quarter of the chum fry typically display flared gills and may rest near the bottom when they first encounter saltwater. This response may last ten to twenty minutes before the fry regain normal swimming activity, and potentially renders them more vulnerable to predators.

## Results

Cowling Creek Hatchery chum scale data can be used to determine adult spawner age ratios (Table 1). Although age 4 adults predominate in the run years observed, no consistent pattern is apparent because the numbers of adults returning each year varies. The age of the adult spawners can be used to generate survival

of each brood year (Figure 4). This data set displays the trend for Cowling Creek chum to return in higher proportion as age 4 adults. The average survival to the hatchery rack was 0.5% for the years 1977 to 1989. This survival to rack rate will increase significantly when the age data for brood years 1990 to 1995 is available. Significant non-treaty commercial gillnetting and purse seining occurred from 1987 until 1993 outside of Miller Bay and may have harvested up to half the returning adult chum. The decrease in commercial value for chum salmon resulted in very reduced non-treaty fishing effort after 1993 and may explain the increased hatchery return. No Cowling Creek chum salmon are tagged.

## Results

Cowling Creek Hatchery satellite chum eggbox releases have stabilized or increased escapement to Dogfish, Big Scandia, and Barker Creeks even with directed terminal fisheries on these streams (Figure 5). Chico Creek chum reflect primarily wild escapement, but one of its tributaries, Dickerson Creek, has a substantial eggbox component to its escapement. Dickerson Creek had two blocking culverts rendering it impassable to chum salmon for decades. There were few spawning adult chum in Dickerson Creek even after fish ladders were installed in the early 1980's. The first significant Dickerson Creek chum returns coincided with the expected returns from the eggbox releases. The majority of the chum returning to Dickerson Creek in the mid to late 1980's displayed the same behavior observed below the Dogfish, Big Scandia, and Barker Creek eggboxes: the adults attempt to swim up the eggbox water source instead of staying in the main stream channel. The main streams generally have flows 25-50 times greater than the eggbox tributaries. Most of the chum will finally spawn in the main stem instead of the tributary.

The other satellite eggbox sites have smaller releases but show similar patterns except Clear Creek. Clear Creek, located near Silverdale, has had consistently low survival from two chum eggbox release sites. Chinook and coho reared in Clear Creek also have very low survival, and natural salmon spawning is almost nonexistent. Possible explanations include pollution, but exact causes are unknown at this time.

Survival of the satellite eggbox unfed volitional releases is estimated to be 0.1 to 0.2% back to the stream.

## Chum Salmon Enhancement

Increased eggbox production since the mid-1980's have coincided with an increased commercial catch (Figure 6; also see Figure 7 for the value of the Tribal catch). This relationship may not be significant overall because east Kitsap's primary chum production is wild Chico stock. The relationship is significant in Liberty Bay, fed by Big Scandia and Dogfish Creeks, because a Tribal chum fishery has been reestablished for the first time in decades.

Orca predation had a significant impact on the Chico Creek run in 1997 by consuming an estimated 20,000 adult chum. Escapement into Chico Creek for 1997 will probably be less than 5,000 adults, below the desired escapement of 16,000 to 18,000 adults. Orca had not been observed feeding upon Chico chum for four decades. If the Orca return more frequently, their impact to the wild Chico chum population could be significant given the urbanizing watershed.

All of the streams with eggboxes are presently undergoing projects related to restoration and barrier removal. The Boy Scouts, sports groups, tribes, local cities, county government, WDFW, USFWS, and other agencies are involved, and these projects will probably increase with the current emphasis on the Wild Salmonid Policy. The eggboxes were originally intended to "seed" streams, then be removed as chum salmon management is based on natural production. However, Kitsap County's urbanization rate has been rapid and the impacts are quickly felt within the small watersheds. The Tribe is presently evaluating the costs and benefits of a longer-term eggbox program as well as implementing a feeding strategy to increase the survival rate of selected enhanced chum populations.



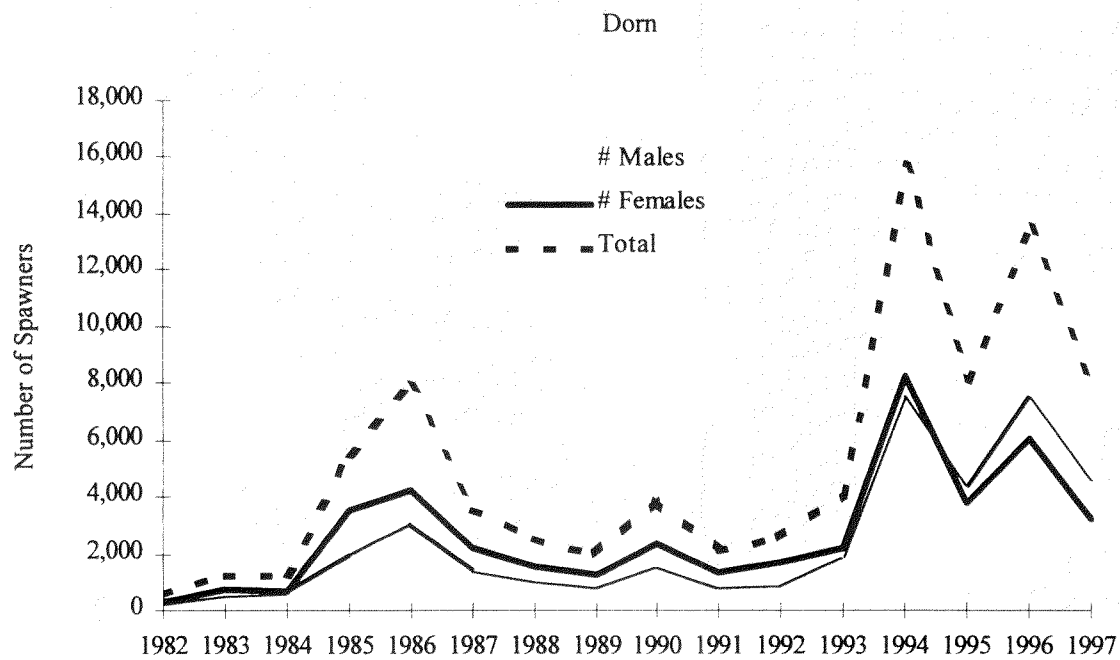


Figure 2. Cowling Creek Hatchery adult chum salmon return by sex, 1982 to 1997.

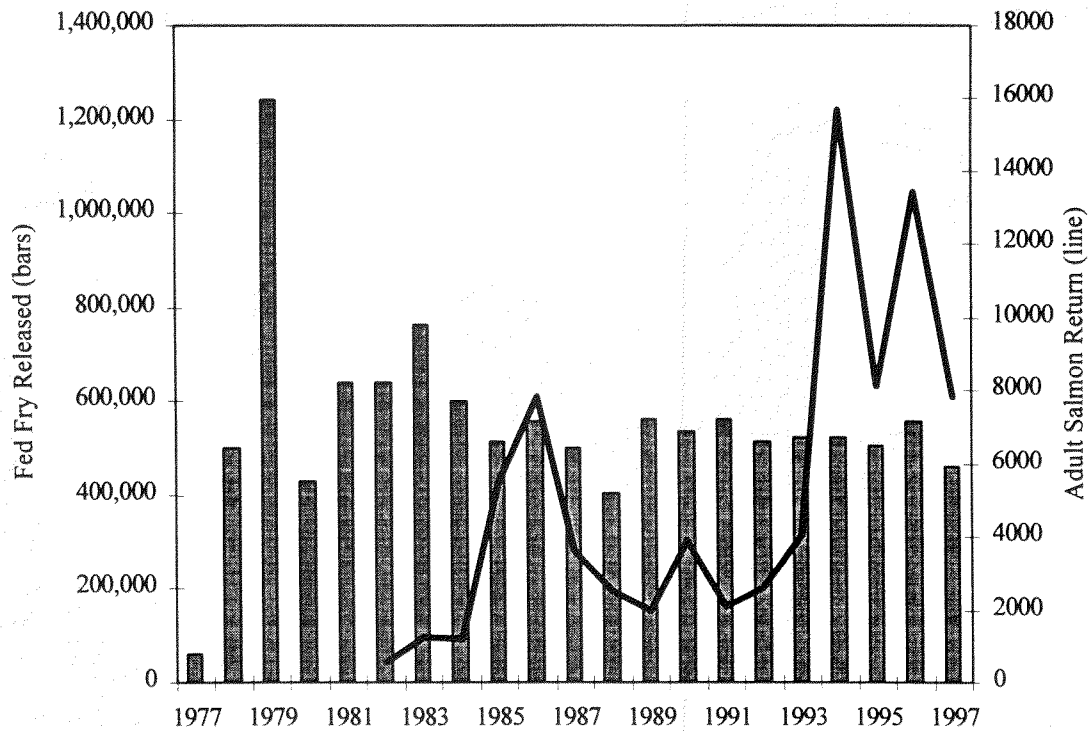


Figure 3. Cowling Creek Hatchery fed fry release and adult salmon return, 1977-1997.

# Chum Salmon Enhancement

Run Year	% Age 3	% Age 4	% Age 5
1980	100	0	0
1981	13	87	0
1982	27	72	1
1983	48	41	11
1984	41	56	3
1985	82	17	1
1986	24	74	2
1987	27	67	6
1988	35	58	7
1989	45	52	3
1990	4	93	3
1991	47	47	6
1992	10	84	6
1993	63	25	10
1994	17	81	2

Table 1. Cowling Creek Hatchery chum age by run year, 1980 to 1994.

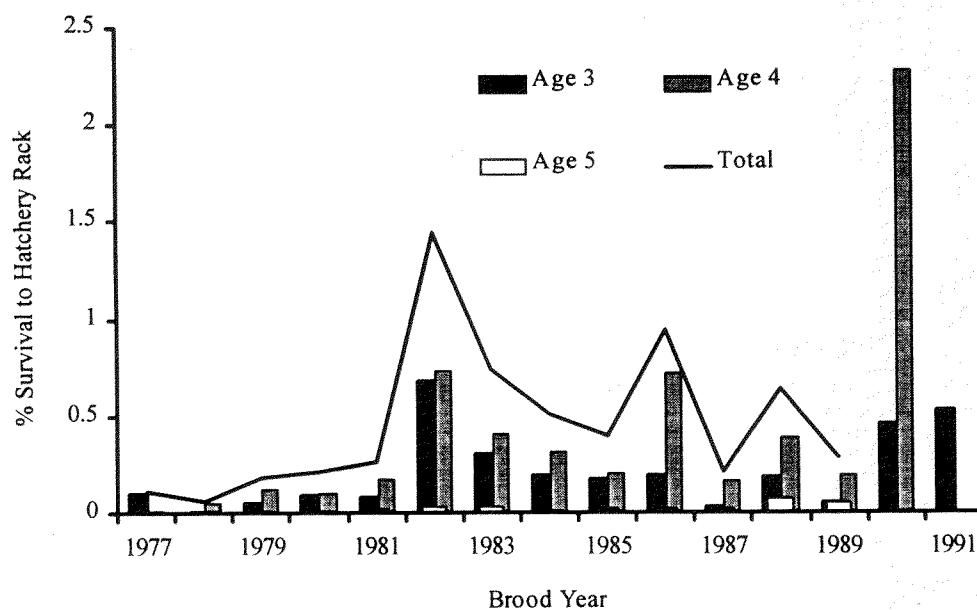


Figure 1. Cowling Creek Hatchery percent survival to hatchery rack by brood year, 1977 to 1991.

# Dorn

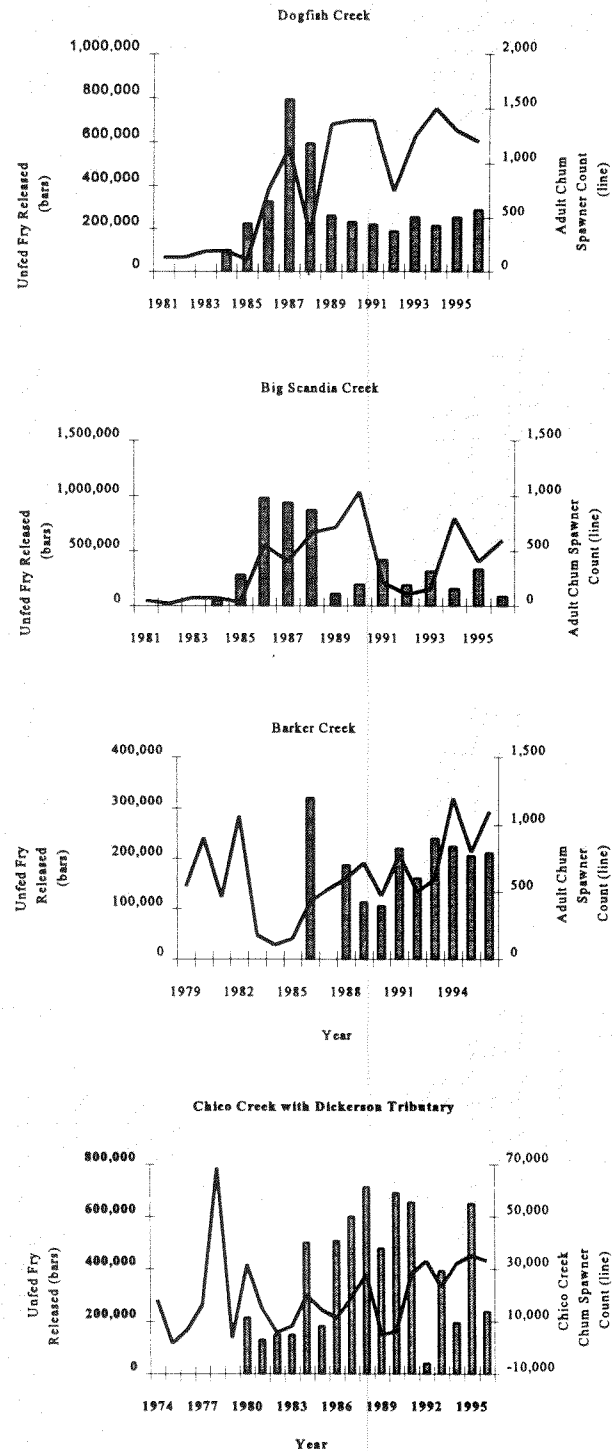


Figure 1. Unfed fry eggbox releases and adult spawner counts.

# Chum Salmon Enhancement

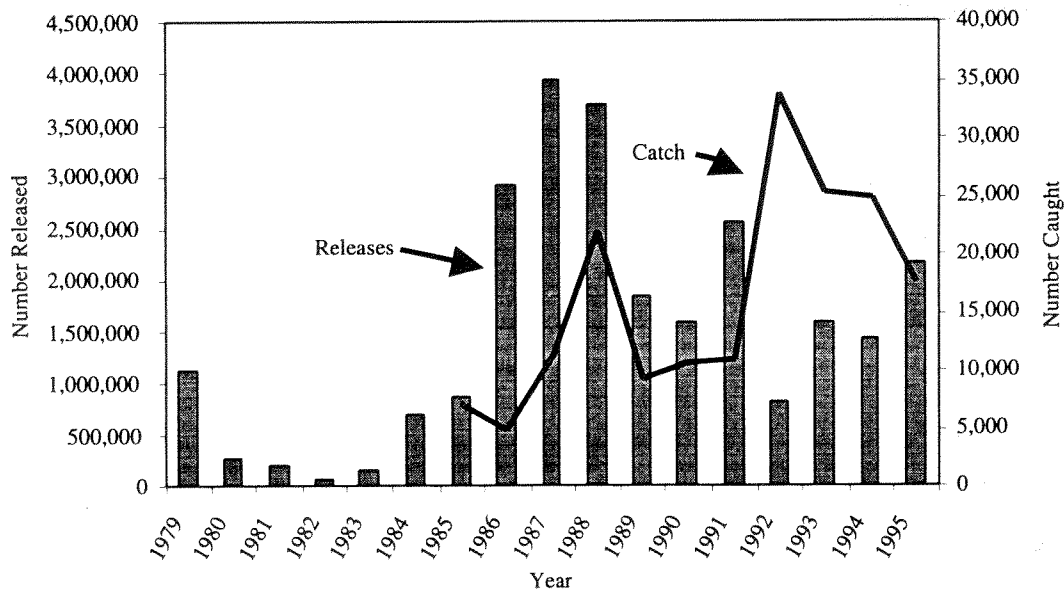


Figure 6. Suquamish Tribe Area 10E commercial chum harvest and East Kitsap chum enhancement.

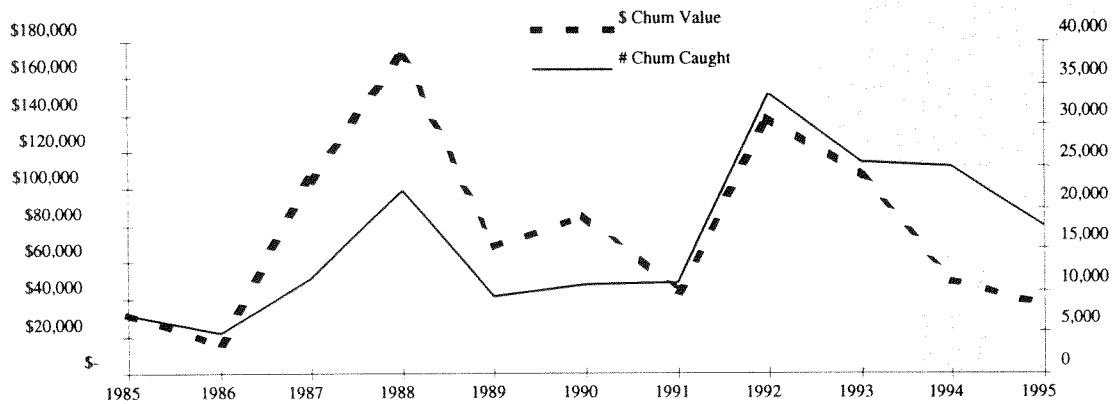


Figure 7. Suquamish Tribe Area 10E commercial chum harvest and value.

Dorn

# Session III

## **New Natural Rearing Practices**

Session Chair:

**Dr. Conrad Mahnken**  
(National Marine Fisheries Service)

# Predator Avoidance Training Can Increase Post-release Survival of Chinook Salmon

Desmond J. Maynard, Anita LaRae, Gail C. McDowell,  
Glen A. Snell, Thomas A. Flagg & Conrad V. W. Mahnken  
*Resource Enhancement and Utilization Technology Division  
Northwest Fisheries Science Center  
National Marine Fisheries Service  
National Oceanic and Atmospheric Administration  
2725 Montlake Boulevard East  
Seattle, Washington 98112*

**Abstract** — Predator avoidance training may be a tool fish culturists can use to increase the post-release survival of hatchery-reared salmonids. Laboratory studies indicate salmonids observing predation on conspecifics have a higher probability of survival in subsequent predation challenges than predator naive fish. In order to test this concept on a hatchery scale, we stocked 16,000 fall chinook salmon (*Oncorhynchus tshawytscha*) swimup fry into each of six 6,000 liter fiberglass raceways equipped with predator tight covers. Fish in three raceways were designated as controls and prior to release were never exposed to predacious birds or fish. Salmon in the other three raceways were exposed to great blue heron (*Ardea herodias*), hooded merganser (*Lophodytes cucullatus*), largemouth bass (*Micropterus salmoides*), and brown catfish (*Ictalurus nebulosus*) predation prior to release. After exposure, tagged fish from each raceway were released into a Puget Sound tributary stream, Curley Creek, to evaluate the effect of training on post-release survival. Significantly ( $P < 0.05$ ) more trained than untrained chinook salmon were recovered at a downstream weir. The 26% higher relative recovery of trained versus untrained fish suggests enhancement and conservation hatcheries can use this approach to increase salmon post-release survival.

## Introduction

Fish culturists may be able to use predator avoidance training to improve the post-release survival of hatchery-reared salmonids (Maynard et al. 1995). Laboratory studies indicate salmon rapidly learn to recognize and avoid predators after observing attacks on conspecifics (Patten 1977, Thompson 1966, Olla and Davis 1989). This predator recognition increases an experienced fish's chance of surviving during subsequent predator encounters. Research has also demonstrated that conditioning chinook and chum salmon to avoid electrified models of predacious rainbow trout increases their survival in natural and artificial streams (Thompson 1966, Kanayama 1968). The experiment described in this paper determines if conditioning salmon to avoid live predators also increases post-release survival.

## Methods

Ninety-six thousand fall chinook salmon swimup fry donated by the Washington Department of Fish and Wildlife (WDFW) Minter Creek Hatchery were transported to the National Marine Fisheries Service (NMFS) Manchester Marine Experimental Station where they were systematically divided into six equivalent lots. Each lot was then ponded into one of six pilot scale raceways (6.4 m long by 1.5 m wide with a 0.6 m water depth) located at the laboratory's freshwater fish culture

facility. The fish in three raceways received experimental predator avoidance training, while fish in the other three raceways served as untrained controls. The control raceways were always covered with bird-tight netting to ensure that unintended predator exposure did not confound the results. Except for predator avoidance training, the fish in both treatments received identical husbandry and were reared following standard salmon culture protocols.

The training process employed a diverse array of predators to ensure the fish were exposed to at least one species they would encounter after release. This also provided us an opportunity to compare each predator species' suitability for conditioning avoidance behavior in hatchery-reared salmon.

In March 1997, training was initiated by uncovering the three predator avoidance conditioning raceways to allow local fish-eating birds access to the fish. Although a young great blue heron (*Ardea herodias*) occasionally fished the raceways, it disappeared within a few weeks and was not observed again. Belted kingfishers (*Ceryle alcyon*) occasionally flew overhead during the study, but were never observed to fish in the raceways. Therefore, we considered this insitu predator exposure at best a limited event.

The primary predator training sessions were conducted by placing predacious birds and fish in cages placed in the

raceways. The cages were constructed of a 1.6 m long by 1.1 m wide by 1.1 m tall polyvinyl chloride (PVC) pipe frame that was completely covered with a 3.8 by 3.8 cm mesh net. This size mesh allowed chinook salmon fry to freely swim in and out of the cage, while confining larger predacious birds and fish within the cage. When they were placed in the raceways, the top half of the cages were suspended above the water so that piscivorous birds would not drown. Cages containing no predators were frequently placed in the raceways so that the fish would learn to associate predation events with predators, rather than presence of the cage.

Two phases of cage training were conducted. First, hooded mergansers (*Lophodytes cucullatus*) were placed into these cages for seven 50 minute long training periods in late April 1997. In nearly all training sessions, the mergansers were removed from the raceway before they ceased fishing. This ensured salmon fry experienced nearly continuous negative reinforcement from these predators. The next training experience involved placing two largemouth bass (*Micropterus salmoides*) and one brown catfish (*Lophodytes cucullatus*) in each cage for a week. Prior to being placed in the cage, each fish was tested to ensure it ate chinook salmon fry. Both types of cage training experiences were completed by mid-May 1997.

The effect of predator avoidance training on post-release survival was evaluated with releases of study fish into the Curley Creek watershed in Kitsap County, WA. These releases were conducted with representative samples of fish from each of the six raceways. These sample fish were removed from the raceways, transferred to six 1.5 m diameter circular tanks, and held from the end of May 1997 until they were released in July 1997. In June 1997, about three weeks before the first release was initiated, the fish were measured to the nearest mm, weighed to the nearest 0.1 g, and tagged with passive integrated transponders (PIT tags). The salmon in the three circulars from the predator avoidance conditioning raceways had their training reinforced by placing one largemouth bass in each tank for the nights of 25 June and 30 June 1997. The unconstrained predators were allowed to prey upon chinook salmon fry in the circular tanks overnight and were removed early the next day.

Releases began three days after the last retraining session, with 51 fish being trucked and released into each of two Curley Creek tributaries on 3 July 1997. The release site on each tributary was 1.3 km upstream of our smolt collection weir on Curley Creek. We were concerned that

contagious behavior might confound the results. Contagious behavior is a form of social learning where naive animals mimic the behavior that more experienced members of their group display to predators, food sources, and other new stimuli. We minimized this possibility of contagious behavior confounding the results of the study by releasing fish from only one rearing treatment in each tributary on a given release day. The possibility of fish from the two rearing treatments meeting each other at the release sites was further reduced by allowing at least 48 hours to pass between releases. Tributary effects were controlled by alternating the tributary the fish were released into from one release to the next. A total of 511 control and 510 predator trained fish were released into the Curley Creek watershed during the 10 releases. The difference in recovery between the two treatments was compared with contingency table analysis.

## Results

In our study, chinook salmon rapidly learned to avoid mergansers. Prior to the introduction of mergansers to a training cage placed in a raceway, fry readily swam into and out of the cage. However, after three training sessions with mergansers, few fry continued to enter the cage. By the fifth session, almost no chinook salmon entered the cage and nearly all the fish remained at least 15 cm from the cage. Initially, the mergansers averaged more than nine prey per training session. However, this average rapidly declined to less than six prey per training session as fry became conditioned to avoid the birds.

The predator avoidance behavior induced in chinook salmon by largemouth bass and brown catfish differed from that induced by mergansers. When bass and catfish were first introduced, few chinook salmon fry entered the cage. However, chinook salmon began to enter the cage within a day, and after a week's residence with these piscivorous fish there were as many chinook salmon in the cage as outside. This change in prey distribution over time may be related to the difference in merganser and bass hunting tactics. Unlike mergansers, bass and catfish did not continuously pursue prey. Instead, they passed their time either holding in place or slowly cruising around the cage perimeter. Although all fish used in training were proven predators, their appetites were not as great as the mergansers'. For instance, the largemouth bass used in reinforcement training in the circular tanks averaged only five chinook salmon during the 17 hour overnight training period.



Training did not appear to affect fish growth. At tagging, the average fork length (Figure 1) of fish in the trained and control treatments did not significantly ( $P = 0.702$ ) differ. The weight (Figure 2) of fish from both treatments also did not significantly ( $P = 0.110$ ) differ at tagging.

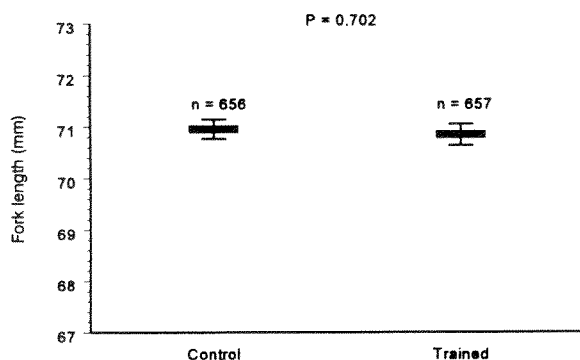


Figure 1. Average fork length of control and predator avoidance trained fall chinook salmon. Horizontal bars are mean values and vertical bars are standard error.

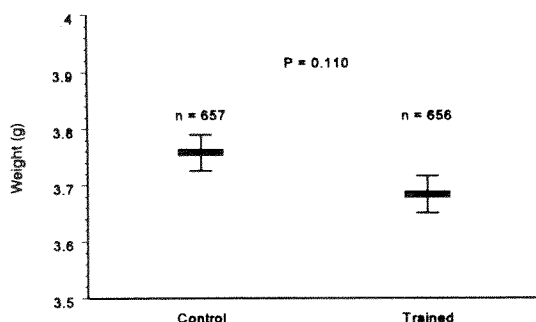


Figure 2. Average weight of control and predator avoidance trained fall chinook salmon. Horizontal bars are mean values and vertical bars are standard error.

Predator avoidance training appears to have increased chinook salmon post-release survival in our study. The post-release recovery of predator conditioned fish was significantly ( $P = 0.046$ ) higher than that of control fish (Figure 3). The relative survival  $[(\% \text{ recovery experimental treatment} - \% \text{ recovery control treatment}) / (\% \text{ recovery control treatment})][100\%]$  of predator conditioned fish was 26% higher than that of control fish. Within a week of the last release, the recovery rate of fish from both treatments had drastically dropped. Although

the weir was operated into September, only 18.6% of all the fish released were recovered.

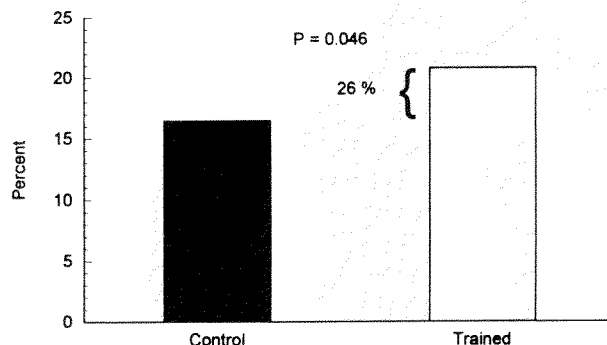


Figure 3. Percent post-release recovery of control and predator avoidance trained fall chinook salmon recaptured at the Curley Creek weir.

## Discussion

This study confirms that predator avoidance training with live predators can increase the post-release survival of hatchery-reared salmonids. The benefits of this training can be considerable with the post-release survival of predator trained fish being 26% higher than untrained fish. The predator avoidance training protocol used in this study required only a slight increase in operational costs. During predator avoidance training, less than two hours of personnel time are expended per day in handling mergansers. The birds rapidly learned to enter the training cage, the cage was easily transported to and from the raceways, and once the cage was placed in the raceway it did not appear to interfere with routine fish culture operations. A pair of hooded mergansers can be purchased for about \$125, and it takes less than 10 minutes a day to maintain them in captivity. The bass and catfish have similarly low acquisition, handling, and maintenance costs. The increased survival benefits of predator avoidance training thus far outweighed the slight increase in operational costs in our experiment.

Predator avoidance training has a very favorable cost:benefit ratio. This ratio is based on the number of fish sacrificed in training that would have successfully migrated downstream compared to the increase in number of successful downstream migrants due to training. The prerelease exposure of chinook salmon to limited (0.6% mortality) hooded merganser and largemouth bass predation increased post-release survival by 26%. This required that approximately 100 fish be sacrificed to train the remaining 15,900 fish to avoid predators after release. With the 20-50% instream survival rates experienced by

control fish in past studies (Maynard et al. 1995), this predator training produces an additional 775-1937 fish surviving migration through the stream corridor. This yields a very favorable 1:40 cost:benefit ratio.

Although predator avoidance training is a useful tool for increasing post-release survival, it only needs to be implemented at those facilities that produce predator naive fish. Hatcheries allowing predators to enter their ponds due to a lack of bird netting and electric fences are probably already providing uncontrolled predator avoidance training.

Programs using hatcheries to produce fish to enhance the fishery or mitigate for habitat loss can potentially derive several benefits from adopting predator avoidance training protocols. The most obvious benefit would be to simply use the increased post-release survival generated by predator avoidance training to boost the number of fish available for harvest. The increased post-release survival generated by predator avoidance training might also be used to reduce the number of fish that must be reared and released to produce an equivalent number of fish for harvest or to meet mitigation goals. The increased survival generated by predator avoidance training could be used to lower operational costs with fewer fish needing to be fed, marked, etc. to produce an equivalent number of recruits to the fishery. Increased post-release survival would permit facilities to meet their enhancement and mitigation goals, while removing fewer wild fish for broodstock and releasing fewer smolts to negatively interact with wild fish in the migratory corridor. Both these factors are important considerations in permitting enhancement operations to continue in areas where they may impact endangered and threatened stocks.

The development of predator training protocols is in its infancy. Research should be conducted to determine if live predators or electrified models (such as those used by Thompson 1966 and Kanayama 1968) provide the best conditioning stimulus. Work should also be carried out to determine if visual, acoustic, chemical, or a combination of cues provides the information necessary for effective predator avoidance training. This research will not only refine techniques, but will provide nonlethal training protocols that can be used in the reintroduction of endangered and threatened stocks of salmon.

Predator avoidance training offers conservation programs an urgently needed opportunity to increase the survival of captive reared animals that are being reintroduced to the wild. In supplementation programs, where a small number

of fish may be sacrificed in training, live predators can be used to condition fish to avoid predators they will encounter after release. However, at facilities rearing protected fish that cannot be sacrificed during training, nonlethal approaches to predator avoidance training probably should be used. Nonlethal training may potentially be accomplished by conditioning fish with electrified predator models. Alternatively, if visual cues are all that is needed to condition fish to avoid predators, nonlethal training may be achieved by simply having captive-reared fish visually witness (live or videotaped) predation events on conspecifics from nonlisted stocks. In general, predator avoidance training is a valuable technique that both fishery enhancement and conservation hatcheries can use to increase the post-release survival of their fish.

### References

- Kanayama, Y. 1968. Studies of the conditioned reflex in lower vertebrates: X. Defensive conditioned reflex of chum salmon fry in group. *Mar. Biol.* 2:77-87.
- Maynard, D. J., T. A. Flagg, and C. V. W. Mahnken. 1995. A review of semi-natural culture strategies for enhancing the post-release survival of anadromous salmonids. *Am. Fish. Soc. Symp.* 15:307-314.
- Olla, B. L., and M. W. Davis. 1989. The role of learning and stress in predator avoidance of hatchery-reared coho salmon (*Oncorhynchus kisutch*) juveniles. *Aquaculture* 76:209-214.
- Patten, B. G. 1977. Body size and learned avoidance as factors affecting predation on coho salmon (*Oncorhynchus kisutch*) fry by torrent sculpin (*Cottus rhotheus*). *Fish. Bull.* 75:457-459.
- Thompson, R. B. 1966. Effects of predator avoidance conditioning on the postrelease survival rate of artificially propagated salmon. Ph.D. Thesis, Univ. Washington, Seattle, 155 p.

# Chemical Alarm Signaling in Chinook Salmon Smolts: An Opportunity for Anti-predator Conditioning

Barry A. Berejikian, E. Paul (Skip) Tezak, and Thomas A. Flagg

National Marine Fisheries Service, Northwest Fisheries Science Center, Resource Enhancement and Utilization  
Technologies Division, 2725 Montlake Blvd. East, Seattle, Washington 98112-2097, USA,

R. Jan F. Smith

Dept. of Zoology, University of Saskatchewan, Saskatoon, SK, Canada S7N 5E2

Steve L. Schroder and Curtis M. Knudsen

Washington Department of Fish and Wildlife, 600 Capitol Way N., Olympia, WA 98501

**Abstract** — Anti-predator conditioning provides potential to increase postrelease survival of hatchery-reared Pacific salmon (*Oncorhynchus* sp.). Several laboratory studies have demonstrated that juvenile salmonids can be conditioned rather quickly to recognize and avoid predators on the basis of visual cues. Evidence also exists that numerous non-salmonid fish species and rainbow trout (*O. mykiss*) communicate danger through chemical alarm pheromones contained in epidermal cells. When these cells are ruptured and the pheromone is released (e.g., during an attack by a predator), conspecifics exhibit a fright response, and some species can learn to associate the predator odor with danger. Our initial laboratory studies indicated that chinook salmon smolts also communicated danger through chemical signals and demonstrated some capacity to exhibit a learned behavioral response to the odor of predatory cutthroat trout (*O. clarki*) that was previously paired with the odor of an injured conspecific. However, preliminary results from our initial attempts to condition fish with paired alarm signals in hatchery vessels did not indicate that learning (measured by anti-predator behavior) occurred. If effective, the application of paired chemical alarm signals prior to release would be a non-intrusive, relatively simple method that could improve predator recognition, and ultimately increase survival after release.

## Introduction

The postrelease survival of hatchery-reared juvenile Pacific salmon (*Oncorhynchus* sp.) largely depends on their ability to avoid predation. Laboratory studies have demonstrated that juvenile salmon can learn to avoid predators (Patten 1977, Olla and Davis 1989, Berejikian 1995), but in-culture predator training efforts have had limited success in improving survival of juvenile salmonids released into natural streams (see Thompson 1966, Kanayama 1968). The usefulness of predator training programs will depend on their ability to significantly improve postrelease survival and whether they can be easily and inexpensively applied to existing hatcheries.

Numerous non-salmonid fish species and rainbow trout (*O. mykiss*) communicate danger through chemical alarm substances (pheromones) contained in their epidermal cells (Smith 1992, Brown and Smith 1997, Brown and Smith in press). When these cells are ruptured and the pheromone is released (e.g., during an attack by a predator), nearby conspecifics become alarmed by the pheromone and exhibit a fright response. Some species can learn to associate the predator odor with the perceived danger and subsequently respond to the predator odor by itself. This suggests the possibility that these chemical pheromones may be used to condition hatchery fish to avoid predators.

We conducted a laboratory experiment to determine whether chinook salmon (*O. tshawytscha*) possess a chemical alarm signaling mechanism, and whether they could be conditioned in laboratory aquaria to recognize and exhibit a fright response to the odor of a predatory cutthroat trout (*O. clarki*). We conducted a second experiment to determine whether introducing a paired alarm signal (conspecific odor and predator odor) directly into hatchery rearing vessels would condition a fright response when the same fish later received predator odor in laboratory aquaria.

## Methods and Materials

### Study Population

Juvenile chinook salmon were obtained from the Washington Department of Fish and Wildlife Bingham Creek Hatchery population. Bingham Creek is a tributary to the East Fork Satsop River, which flows into the Chehalis River and eventually into the Pacific Ocean at Grays Harbor, WA. One thousand emergent chinook salmon fry were stocked from incubation trays into each of five, 1.8-m diameter vessels at the Bingham Creek Hatchery on 28 February 1997. The fish were fed a standard commercial salmon diet several times daily, 5 days per week.

## Experiment 1 -

### *Fish Treated in Aquaria Stimuli*

Eight juvenile chinook salmon were used to generate the skin-muscle extract. The salmon were killed with a blow to the head. A total of 30 cm<sup>2</sup> of skin and muscle tissue were removed from the eight chinook salmon. The tissue was added to 1,500 ml of distilled water, homogenized and filtered through a polyester filter floss. The procedure was repeated to create a "control" extract from 12 swordtails (*Xiphophorus helleri*), which possess no alarm pheromone. The extracts were frozen in 50 ml lots. Cutthroat trout odor was produced by holding two cutthroat trout (245 cm and 295 cm fork length) for 15 hours in a 28 L water bath supplied with air. The cutthroat trout were then removed and the water was frozen in 50 ml lots.

### *Protocols*

Fish for this experiment were transferred from the Bingham Creek Hatchery to the NMFS Manchester Marine Experimental Station, Manchester, WA on 1 June 1997 and held in a 1.1-m diameter rearing vessel. Trials were conducted in two nearly identical indoor flumes, each measuring 9.0-m long by 1.5-m wide. Ten, 170-L aquaria were situated in 1 of the flumes and 5 aquaria in the other. The clear side of each aquarium faced the side walls of the flumes, which were constructed of double paned glass and allowed complete viewing of fish in each aquarium. Each aquarium contained a 3- to 4-cm deep layer of 1.0- to 1.5-cm diameter gravel. Water was delivered into each aquarium at a rate of 6 L/minute through a funnel, connected to a poly-vinyl tube that terminated at mid-water depth at one end of the aquarium. Water exited each aquarium through a double siphon at the end opposite the water inflow, such that the water depth was maintained at 25 cm. The water exiting through the siphons entered the flumes and created a fairly constant temperature (12.5 to 13.0 °C) water bath around the aquaria. Light was provided by a solid bank of wide-spectrum florescent lights on a simulated photoperiod of 16L:8D. In the center of each aquarium, a 15-cm by 15-cm square tile was situated on 5-cm tall glass legs to provide overhead cover. A plastic ring (2.5-cm high, 1-cm thick, and 18-cm diameter) surrounded the legs of the tile cover to provide lateral cover.

One chinook salmon was placed into each of the aquaria on 9 June 1997 and fed three times daily over the next 3

days. On 12 June 1997 the fish were each fed approximately 20 commercial salmon food pellets between 0745 and 0830 hours. Paired alarm signal trials consisted of 8-minute pre-stimulus and 8-minute post-stimulus observations. Twenty minutes prior to beginning pre-stimulus observations, we introduced 2.5 ml of live *Daphnia*. At the end of the 8 minute pre-stimulus observation, 50 ml of either chinook salmon extract or swordtail extract (control) combined with 50 ml of cutthroat trout water were introduced through the water inflow tube. During both pre and post-stimulus observations, we recorded: 1) the number of food strikes; 2) the amount of time spent in the lower, middle, and upper thirds of the water column; 3) the amount of time spent under cover; and 4) the amount of time spent motionless.

Two days after the initial trials, the same fish were tested for their response to cutthroat trout odor alone to test the null hypothesis that no acquired recognition learning occurred as a result of the paired stimulus introduction. These trials were conducted in the same manner as the paired stimulus trials except that only cutthroat trout water was introduced after the 8 minute pre-stimulus observations. After the trials were completed, the fish were removed and held individually in 6-cm diameter by 25-cm long tubes, constructed of plastic screening which allowed water to pass through. The 15 individually labeled fish tubes were partially submerged in a common 1.1-m diameter tank. These fish were each reintroduced into their original aquarium 4 days later, and underwent a 2 day re-acclimation before being tested again for their response to cutthroat odor only. Thus, each of the 15 fish was tested for its response to a paired stimulus on 12 June (day 1), cutthroat trout odor only on 14 June (day 3), and cutthroat trout odor only a second time on 21 June (day 10). A second set of 15 trials was conducted such that fish were tested for their response to the paired stimulus on 17 June (day 1), and cutthroat odor only on 19 June (day 3), and again on 26 June 26 (day 10).

## Experiment 2 -

### *Fish Treated in Rearing Vessels Stimuli*

Chinook salmon extract was generated by homogenizing 48 cm<sup>2</sup> of skin and muscle fillet in 1 L of distilled water, filtering the liquid, then diluting it to create a 4 L volume. Cutthroat trout odor was generated by placing two live cutthroat trout (380 mm and 255 mm FL) into 12 L of aerated water for 5 hours.

## Chemical Alarm Signaling in Chinook Salmon Smolts

### Protocols

The experiment was conducted to test the null hypothesis that the behavioral fright responses to the neutral stimulus (cutthroat trout odor) would be the same for chinook salmon smolts treated with a paired alarm signal (chinook salmon extract plus cutthroat trout odor) and those treated with a distilled water control, applied directly to the rearing vessels. On 2 July 1997, two of four 1.8-m diameter rearing tanks received 1 L each of the chinook extract and cutthroat trout odor, and the other two tanks received 2 L of the distilled water control. Fifty fish (4.0 to 5.0 g) from each tank were transported to Manchester by tank truck. Members of each tank were kept separate in 40-L plastic totes with mesh sides that allowed water to pass through. The totes were placed into a common 1.1-L tank, and the fish were fed daily until the experiment began. On 7 July 1997, eight of the aquaria received fish from the tanks receiving a paired alarm signal, and seven aquaria received fish from the control tanks. After 3 days of acclimation in the aquaria, the fish were tested for their response to cutthroat trout odor using the same protocols as in experiment 1. A second round of trials, beginning with acclimation on 11 July 1997, was conducted with eight fish from the control tanks and seven from the tanks receiving a paired alarm signal. Thus, there were a total of 30 trials (15 per treatment)

### Statistical Analyses

We calculated the difference between pre-and post-stimulus durations for the following behaviors: motionless, under cover, and lower third of the water column. For the frequency of food strikes, we calculated the post-stimulus/pre-stimulus value as the response variable to correct for individual variation in foraging rates. The differences in these response variables between the experimental (chinook extract and cutthroat trout odor) and control trials (either swordtail extract and cutthroat trout odor, or distilled water) were compared by Mann-Whitney-U tests.

### Results

#### Experiment 1 -

##### *Fish Treated in Aquaria: Paired stimulus trials*

Chinook salmon smolts receiving a combination of chinook salmon extract (CSE) and cutthroat trout odor (CTO) on day 1 responded by reducing their foraging activity following the stimulus introduction to a greater

extent than did smolts receiving swordtail extract (STE) and CTO ( $P < 0.01$ ; Fig. 1). Fish treated with CSE + CTO also spent significantly more time in the lower third of the water column ( $P = 0.03$ ; Fig. 2), and more time motionless ( $P < 0.01$ ; Fig. 3) after the stimulus was introduced than did STE + CTO-treated fish. The two groups of fish did not differ in their time spent under cover ( $P > 0.50$ ). Thus, fish receiving conspecific extract demonstrated stronger anti-predator responses than those receiving the swordtail extract control.

#### *Cutthroat trout odor trials*

When cutthroat trout odor was provided as a lone stimulus on day 3 to the same fish that received either the paired alarm substance (CSE + CTO) or the control substance (STE + CTO), CSE + CTO-treated fish spent more time motionless than STE + CTO-treated fish ( $P = 0.02$ ; Fig. 3), suggesting that CSE + CTO-treated fish learned to associate cutthroat trout odor with danger. There were no significant differences found between the two groups for any of the other behaviors ( $P > 0.25$  in all cases). When the same fish were later re-tested for their response to cutthroat odor on day 10, there were no significant differences detected between the STE and CSE groups (Figs. 1, 2 and 3).

#### Experiment 2 -

##### *Fish Treated in Rearing Vessels*

When tested for their behavioral responses to CTO in laboratory aquaria, chinook salmon smolts receiving CSE + CTO did not exhibit significant differences for any of the behaviors from those smolts receiving a distilled water control ( $P > 0.25$  in all cases).

### Discussion

Chinook salmon smolts in this study demonstrated the presence of a chemical alarm signaling mechanism by exhibiting much stronger fright responses to combined conspecific extract and a predator odor than to swordtail extract and predator odor. The reduction in feeding, increased time spent motionless, and increased time spent near the substrate are common anti-predator responses that would likely reduce detection by predators (Olla and Davis 1989, Donnelly and Whoriskey 1993, Gotceitis and Godin 1993). When the cutthroat odor was introduced 2 days later (day 3), the chinook salmon previously treated with chinook salmon extract reduced their swimming

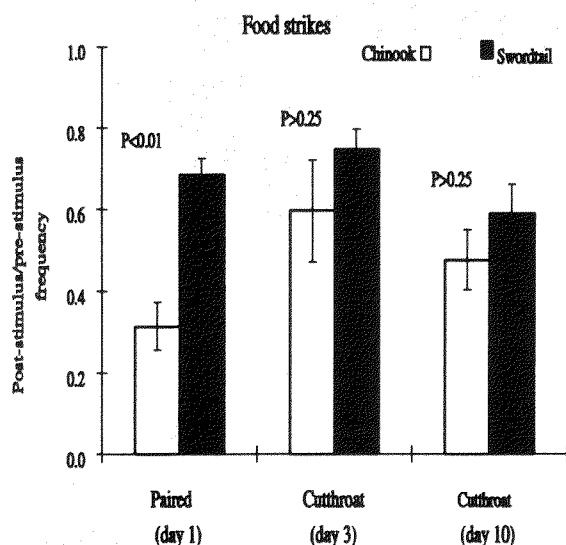


Figure 1. The mean ( $\pm$ s.e) ratio (post-stimulus divided by pre-stimulus) of food strikes made by chinook salmon treated with chinook salmon extract and cutthroat trout odor (test stimulus), compared to those treated with swordtail extract and cutthroat trout odor (control stimulus) on day 1 in laboratory aquaria. Data from the trials on days 3 and 10 represent responses of the fish to cutthroat trout odor only.

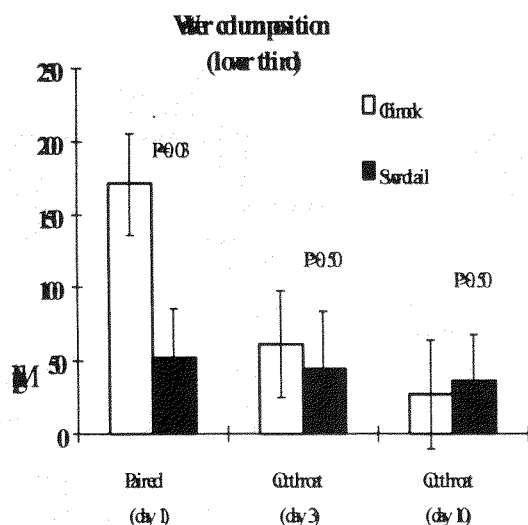


Figure 2. The mean ( $\pm$ s.e) ratio (post-stimulus divided by pre-stimulus) in time spent in the lower <sup>rd</sup> of the water column for chinook salmon treated with the chinook salmon extract and cutthroat trout odor (test stimulus), compared to those treated with swordtail extract and cutthroat trout odor (control stimulus) on day 1 in laboratory aquaria. Data from the trials on days 3 and 10 represent responses of the same fish to cutthroat trout odor only.

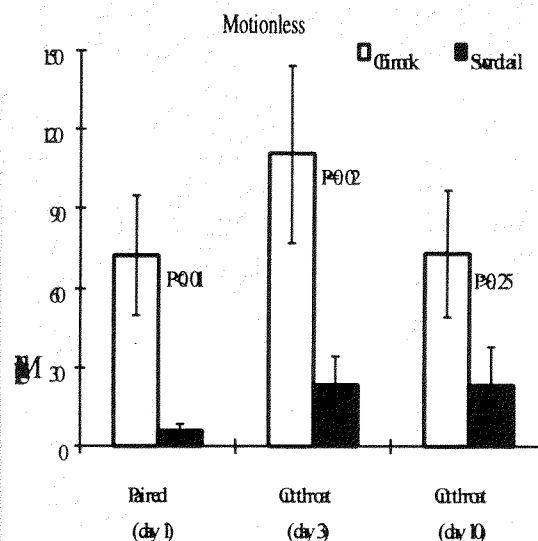


Figure 3. The mean ( $\pm$ s.e) change (post-stimulus minus pre-stimulus) in time spent motionless for chinook salmon treated with chinook salmon extract and cutthroat trout odor (test stimulus), compared to those treated with swordtail extract and cutthroat trout odor (control stimulus) on day 1 in laboratory aquaria. Data from the trials on days 3 and 10 represent responses of the same fish to cutthroat trout odor only.

activity more than those previously treated with swordtail extract. Reduced motion has been demonstrated as perhaps the primary anti-predatory response of chinook salmon (Healy and Reinhardt 1995). This suggests that chinook salmon acquired predator recognition; that is, they learned to associate cutthroat trout odor with danger.

Although there was an indication that learning occurred on day 3 of experiment 1, chinook salmon apparently did not retain the ability to recognize predator odor when tested for their response to cutthroat trout odor 8 days or more after receiving a paired alarm signal in either experiment 1 (day 10 trials) or experiment 2. Among the possible reasons why we observed no significant differences in behavioral responses in those trials is that in both of these experiments the fish were netted from the environment in which they were exposed to the paired alarm signal, then held for several days in small containers, which may have stressed them and interfered with their learning retention. It is also possible that chinook salmon may have a short retention period for this type of learning, although Brown and Smith (in press) demonstrated that rainbow trout were able to retain

acquired predator recognition for at least 21 days. The potentially confounding effects of transport stress (for off-station releases) and learning retention time require further research before anti-predator training with chemical alarm signals is implemented on a production scale.

The effectiveness of chemical stimuli for anti-predator conditioning of chinook salmon depends largely on the strength of their fright response to such cues. The fright responses of chinook salmon (this study, and Healy and Reinhardt 1995) and rainbow trout (Brown and Smith 1997, Brown and Smith in press) to conspecific skin extract appear to have been much less pronounced than those exhibited by a many non-salmonid species (see Smith 1992 for a review), but more pronounced than others (e.g. swordtails: Mathis and Smith 1993). The strongest response we observed was a reduction in swimming activity. Healy and Reinhardt (1995) also noted motionless behavior as the primary defense of chinook salmon against actual predators, suggesting that anti-predator conditioning with paired chemical alarm signals may decrease vulnerability to predators. Whether presenting additional visual or vibrational stimuli during predator training would strengthen predator recognition and anti-predator responses remains unclear (see also Olla and Davis 1989).

We assume that the behavioral responses we observed would improve the chances of survival of chinook salmon encountering predators in the natural environment. However, predator training using chemical alarm signals should also be evaluated by conducting predation bioassays and postrelease survival experiments to better ensure its effectiveness. We have initiated such studies as a portion of this research, but at this time data are still being collected and analyzed. If this type of anti-predator training proves effective, it would likely be more amenable to current hatchery operations than other methods which involve, for example, electrified predator models (Thompson 1966, Kanayama 1968), or introducing predators directly into hatchery raceways.

#### References

- Berejikian, B. A. 1995. The effects of hatchery and wild ancestry and experience on the relative ability of steelhead trout fry (*Oncorhynchus mykiss*) to avoid a benthic predator. *Can. J. Fish. Aquat. Sci.* 52:2076-2082.
- Brown, G.E. and R.J.F. Smith 1997. Conspecific skin extracts elicit antipredator responses in juvenile rainbow trout (*Oncorhynchus mykiss*). *Can. J. Zool.* 75:1916-1922.
- Brown, G.E. and R.J.F. Smith (In Press) Acquired predator recognition in juvenile rainbow trout (*Oncorhynchus mykiss*): conditioning hatchery reared fish to recognize chemical cues of a predator. *Can. J. Fish. Aquat. Sci.*
- Donnelly, W. A. and F. G. Whoriskey Jr. 1993. Transplantation of Atlantic salmon (*Salmo salar*) and crypsis breakdown. In R. J. Gibson and R. E. Cutting (editors), *Production of juvenile Atlantic salmon, Salmo salar, in natural waters*, p. 25-34. *Can. Spec. Publ. Fish. Aquat. Sci.* 118.
- Gotceitas, V. and J-G. J. Godin. 1993. Effects of aerial and in-stream threat of predation on foraging by juvenile Atlantic salmon (*salmo salar*). In R. J. Gibson and R. E. Cutting (editors), *Production of juvenile Atlantic salmon, Salmo salar, in natural waters*, p. 35-41. *Can. Spec. Publ. Fish. Aquat. Sci.* 118.
- Healy, M. C., and U. Reinhardt. 1995. Predator avoidance in naïve and experienced juvenile chinook and coho salmon. *Can. J. Fish. Aquat. Sci.* 52:614-622.
- Kanayama, Y. 1968. Studies of the conditioned reflex in lower vertebrates. X. Defensive conditioned reflex of chum salmon fry in a group. *Mar. Biol.* 2:77-87.
- Mathis, A. and R. J. F. Smith. 1993. Fathead minnows, *Pimephales promelas*, learn to recognize northern pike, *Esox lucius*, as predators on the basis of chemical stimuli from minnows in the pike's diet. *Anim. Behav.* 51:185-201.
- Olla, B. L., and M. W. Davis. 1989. The role of learning and stress in predator avoidance of hatchery-reared coho salmon (*Oncorhynchus kisutch*) juveniles. *Aquaculture* 76: 209-214.
- Patten, B. G. 1977. Body size and learned avoidance as factors affecting predation on coho salmon fry by torrent sculpin (*Cottus rotheus*). *Fish. Bull.* 75: 451-459.
- Smith, R. J. F. 1992. Alarm signals in fishes. *Rev. Fish Biol. Fish.* 2:33-63
- Thompson, R.B. 1966. Effects of predator avoidance conditioning on the postrelease survival rate of artificially propagated salmon. Ph.D. Thesis, Univ. Washington, Seattle, WA. 156 p.





# **Addition of Floating and Bottom Structures to Concrete Raceways at Solduc Hatchery**

Geraldine Vander Haegen & Andrew Appleby

*WDFW Hatcheries Program, 600 Capitol Way North, Olympia, WA, 98501-1091*

## **Introduction**

Many of Washington Department of Fish and Wildlife's (WDFW) hatcheries rear fish from fry to smolts in concrete raceways. There are many practical reasons for this - concrete raceways are durable, relatively easy to clean, fish health concerns can be addressed, and the technology is quickly transferrable between hatcheries. However, because these types of rearing containers are so different from the natural environment, the fitness of the released smolts for life in the wild is being questioned, particularly concerning their ability to survive the downstream migration.

In theory, if we could change the rearing environment, then we may also be able to change a fish's behavioral and physiological response to rearing, hopefully producing a smolt that is better able to survive after release. While rebuilding many of WDFW's hatcheries is impractical, retrofitting them with in-pond structures might be possible if we could find a practical, inexpensive structure that produced a smolt that survived better than normal. Steve Schroeder at WDFW, Desmond Maynard at National Marine Fisheries Service (NMFS), and many others have added single and combinations of treatments to tanks of fish with some evidence that the survival of the treated fish after release is improved at least initially (see Maynard et al. for a review). Our study extends this work by using single treatments on full production sized groups of fish, and compares the survival to adult of treated and untreated fish. In this study, we are adding simple floating or bottom structures to production raceways at Solduc Hatchery in an attempt to improve the quality and post-release survival of coho smolts.

## **Objectives**

We have three specific objectives for this study:

- (1) Determine the feasibility of adding simple floating and bottom structures to concrete ponds.

- (2) Monitor effects of structures on growth and smolt status while the fish are in the ponds.

- (3) Monitor effects of structures on survival to adult and fishery contributions by CWT recoveries.

## **Methods**

Solduc Hatchery is on the Solduc River, about 36 miles upstream of its confluence with the Quillayute River on the northwest coast of Washington's Olympic Peninsula. The hatchery rears about 700,000 coho salmon from the egg stage to release as smolts. The fish are placed in Burroughs ponds in late spring, and released on-station one year later. All of the ponds are covered with bird netting, so the fish are exposed to only an occasional predator. We used six ponds in the study, each with a population of about 97,000 coho, of which 25,000 are coded-wire tagged in each pond. Eight weeks before release, treatments are randomly assigned to the six ponds. Two ponds contain floating structures, two contain bottom structures and two are control ponds. All six ponds are fed the same amount of food from demand feeders. Lengths, weights and smolt status (Prentice et al. 1981) are sampled biweekly until release.

Floating structures were constructed from 8' diameter polypipe rings covered with camouflage netting. Ten were placed in each pond and tethered to the raceway. Bottom structures were made from plastic fish totes with the sides cut away. Eight were placed upside down in each raceway and anchored with sandbags to keep them from floating.

## **Results**

We are now beginning the second year of the study. In the first year, we could only use four ponds because of disease outbreaks in two ponds at the time of tagging. Fish in the study ponds did not experience any disease problems. In the first year, we found no difference in growth between the treatment and control ponds (length:  $F=0.77$ ,  $p>0.5$ ; weight:  $F=1.21$ ,  $p>0.3$ ).

The smolt status of a fish was determined using a ranking system based on criteria describing the physical appearance of the fish. Each fish was assigned a rank based on this system and the ranks of the fish from the four ponds were compared. We found that the fish in the treatment ponds were significantly more smolted than the fish in the control ponds (Table 1,  $Z=-2.53$ ,  $p<0.05$ ).

Table 1: Proportion of parr and transitional fish in each of the study ponds; the rest of the fish were smolts.

Pond	Treatment	Parr or Transitional
1	Floating Structure	24%
2	Floating Structure	28%
3	Control	48%
4	Control	36%

### Discussion

In the first year of this study, the addition of floating structures to raceways at Solduc Hatchery produced some physiological changes in the fish resulting in accelerated smoltification compared with the control fish. While promising, whether this observation will be repeated in the next two years, and whether it will result in better survival cannot be determined until the coded-wire tags are recovered over the next four to five years.

The addition of floating structures to a raceway caused little inconvenience to fish cultural operations, especially when demand feeders were used. Although the floating covers became covered with algae shortly after placing them in the ponds, this did not pose any fish health hazard, and it seemed to enhance the effects we tried to

achieve with the netting, so we did not try to remove the algae. The fish were attracted to the covers immediately, and used them extensively, although not all of the fish could fit under the covers at the same time. The fish in the covered ponds had a noticeably stronger flight response to a passing human or dip net than the fish in the control ponds, presumably because of the added cover.

Initial trials with the bottom structures suggest that they will be less convenient for fish culture because of the necessity for weighing them down. Fish were attracted to the bottom structures, and used them extensively, although not all of the fish could fit under the structures at the same time. Growth and smolt status data will be collected in the spring on 1998.

### Acknowledgments

We thank Rob Allan, Rich Watson, Mike Dunar, Ted Daggett, and Dan Evans for their professional assistance and careful attention to the study ponds at Solduc Hatchery.

### References

- Maynard, D.J., T.A. Flagg, & C.V.W. Mahnken. 1996. Development of a natural rearing system to improve supplemental fish quality, 1991-1995. Progress Report. Bonneville Power Administration Report # DOE/BP-20651-1. 216 pp.
- Prentice, E.F. and 11 coauthors. 1981. Assessment of smoltification and fitness for ocean survival (quality) of chinook and coho salmon and steelhead in the Columbia River and Puget Sound hatcheries. Part I Report for FY 1980-81. Part II Project summary and recommendations (1978-1981). Coastal Zone and Estuarine Studies Technical Report.

# Evaluation of Semi-Natural Rearing for Coho Salmon

Howard Fuss, Jim Byrne, and Charmane Ashbrook  
*Washington Department of Fish and Wildlife, Hatcheries Program,  
600 Capital Way N., Olympia, WA 98501-1091*

## Introduction

In nature, stream dwelling salmonids rear at lower densities than conspecifics reared in hatcheries. The natural stream environment includes substrates of varying size and complexity, overhead cover in the form of vegetation, fallen trees or rootwads, and undercut banks. In contrast, hatchery rearing structures lack complexity because the goal is to produce large numbers of fish relative to the amount of available water flow, facilitate periodic cleaning and provide expedient removal of fish when necessary.

Use of conventional fish rearing practices are thought to reduce the survival of hatchery fish relative to wild fish in several ways. First, hatcheries promote very high survivals from the egg to smolt stage relative to the natural environment. Post-release survival of hatchery reared fish is usually lower than wild fish, possibly because a higher percentage of the hatchery population has not experienced strong selective pressures during rearing. This lack of selective pressure may lead to domestication, a process that selects traits in the population that are conducive to survival in the less rigorous hatchery environment, but not in the natural environment. Secondly, hatchery fish are fed a commercially developed diet that may lack micro-nutrients found in natural feeds that promote better survival. Because the commercial diet is fed in the form of a pellet, the delivery of the pellet to the fish is thought to condition the fish to surface oriented feeding, thus exposing the fish to higher rates of predation from avian predators.

Recently, results of several experiments have illustrated some key differences in physiology between hatchery and wild fish at the time of seaward migration. Shrimpton et. al. (1994) compared saltwater tolerance of coho salmon juveniles that were reared (1) exclusively in the hatchery, (2) hatchery fish transplanted into the upper watershed (colonized), and (3) wild fish of hatchery ancestry. Hatchery fish showed a reduced tolerance to seawater as assessed by a greater perturbation in plasma sodium concentration following transfer to saltwater.

Additionally, hatchery fish showed significant declines in hematocrit, significant increases in circulating plasma cortisol concentration, fewer chloride cells and lower specific activities of the enzymes  $\text{Na}^+ - \text{K}^+$  ATPase and citrate synthase. The authors concluded that the higher mortality of hatchery fish after transfer to saltwater was due to rearing environment, and subsequent survival after entering seawater would be compromised relative to colonized or wild fish. The rearing structure at the hatchery were earthen channels supplied with ground water ( $10^0 \pm 2^0$ ) after it had passed through raceways (reuse water).

Companion studies similarly showed that hatchery fish possessed lower levels of corticosteroid receptor (Shrimpton et al. 1994) and a reduced hypo-osmoregulatory ability in seawater affecting swimming performance relative to wild fish (Brauner et. al. 1994).

Maynard et al (1996) have conducted research on NATURES rearing of chinook salmon to test four underlying assumptions: 1) promotion and development of cryptic coloration and anti-predator behavior; 2) increased post release foraging efficiency; 3) improved fish health and condition by alleviating chronic, artificial rearing habitat-induced stress; and 4) reduction in potential genetic selection pressures induced by the conventional salmon culture environment. To date their research has shown mixed results. For example, chinook reared in NATURES tanks had more natural type body camouflage coloration patterns, a greater fright response to overhead movement, and a 25-50% survival advantage during migration in the stream corridor when released in clear running streams compared conventionally reared groups. However, NATURES rearing had no benefit to fish released into turbid streams, anti-predator training had mixed results with fall chinook, and live food supplementation failed to demonstrate improved instream foraging efficiency of spring chinook.

Rearing environments provided to hatchery fish are based on conventional methods used to maximize production from a given amount of water supplied to the hatchery. The most efficient use of this water is in concrete

raceways or asphalt rearing ponds where large numbers of fish can be reared for a given volume and flow rate of water. For example, typical rearing density for coho salmon ranges from 3.32 kg/m<sup>3</sup> (low) to 36.5 kg/m<sup>3</sup> (high) with normal densities in the range of 4.5-6.5 kg/m<sup>3</sup>. Loading rates typically range in the 1.5-2.5 kg/Lpm (8-12 lb/gpm). The effect of rearing density on survival varies among species. Studies have shown that adult yield of coho salmon is increased with increased rearing density even though percent yield is neutral or slightly impaired. For chinook salmon the reverse is true: increased density both reduces survival and thus adult yield.

Although NATURES technology can be retrofitted to existing hatchery raceway systems, it has not been tested under full production situations, nor has it been demonstrated with species other than chinook. If NATURES rearing can be done at production levels and impart a survival advantage, then increased production of adults over conventional rearing would occur. However, if NATURES rearing requires reductions in the number of fish released to be successful, the fish reared in these converted vessels would have to survive at much higher rates than conventionally reared fish. For example, a hatchery using conventional rearing methods can usually expect a 0.5% survival rate when releasing 1 million chinook smolts, which would produce about 5,000 adults. If it were the case that rearing density in NATURES ponds were to be reduced by 75% (250,000 juveniles released) then to produce 5,000 adults would require a post-release survival of 2%.

### Purpose

The purpose of this study is to determine if a conventional rearing pond can be converted to a semi-natural rearing pond and successfully produce coho smolts. Because of logistical constraints, rigorous scientific design criteria could not be employed, and therefore the study design does not include replication and several variables such as rearing density and loading rates differ among the treatment and control groups. This study incorporates a rearing scenario that could be realistically incorporated in most hatchery operations. The study addresses two questions: 1) Do semi-natural rearing conditions at a hatchery produce smolts that survive at higher levels, and have different smolt characteristics than those reared under standard conditions? 2) If increased survival does occur, is it high

enough to offset losses in production that would occur using the "semi-natural" rearing environment?

Two hypotheses will be tested in the evaluation that will be repeated for three successive broods:

HO<sub>1</sub>: Coho reared in a "semi-natural" pond will have similar Na<sup>+</sup> K<sup>+</sup> ATPase concentrations, and plasma chloride levels, as coho reared in a conventional rearing pond.

HO<sub>2</sub>: Coho reared in a "semi-natural" pond will have similar survivals, as measured by coded wire tag recoveries, as coho reared in a conventional rearing pond.

### Study Protocol

Coho survival on the Columbia River varies annually and ranges from 0.1%-8.0%. Typically coho are reared in conventional asphalt or concrete rearing containers and released as a group over a 1-3 day period, or volitionally over several (2-10) weeks. Most coho rearing programs use surface water and loadings exceeding 1.19 kg fish/Lpm (10 lb fish/gpm) inflow.

The data presented by Shrimpton et al (1994) is compelling because it suggests that conventional rearing of coho produces a high proportion of smolts incapable of survival in seawater. However, several of the study protocols used by those researchers may have affected their results. Specifically, they measured physiological parameters of hatchery fish that were still resident in the rearing pond rather than outmigrating smolts. Also, the hatchery fish were reared on re-use groundwater which has been implicated in impairing smoltification because of its relatively constant temperature. Lastly, they compared hatchery fish reared at high densities, which has been shown to adversely affect the smoltification process.

The study protocol we incorporated for the Elochoman Hatchery did not eliminate all of the above variables that might confound results. For example, fish in the study groups are to be reared at times on re-use surface water, instead of reuse ground water. Also, the fish in both the control and treatment groups will be reared at normal to low densities, and lastly only smolts that migrated from the rearing ponds will be assayed.

## Evaluation of Semi-Natural Rearing for Coho Salmon

### Methods

A single dirt bottomed rearing pond (Semi-natural; Pond 22) (Table 1) at the Elochoman Salmon Hatchery was modified to simulate a natural off-channel rearing pond by adding pit run gravel and large woody debris (LWD) to the pond to provide natural cover. The LWD consisted of small alder tops, fir branches, small logs and stumps. The LWD occupied about 25% of the pond area. The gravel and LWD was added to the pond several weeks before fry were introduced. The pond was filled after the substrates were added, but fry not stocked in the pond until two weeks later.

Coho fry, reared in conventional concrete raceways were added to the treatment pond in June 1996. Thirty one thousand 1995 brood Type N coho fry (average of 4.5 g) were stocked in the treatment pond in mid-June 1996. This pond normally rears about 425,000 coho to release (561 fish/m<sup>2</sup>, 1.6 kg/Lpm). The maximum rearing density and loading rate for the treatment pond with 31,000 smolts was 34 fish/m<sup>2</sup> and 0.09 kg/Lpm, respectively. Coho, from the same source as the treatment pond, were stocked into a dirt bottomed rearing pond (control, pond 21) in early February when they were about 9g. A total of 263,000 fish were stocked in the pond. This pond normally rears about 450,000 coho to release ( 956 fish/m<sup>2</sup>, 0.87 kg/Lpm). The rearing density and loading rate of the control pond at the stocking level used in 1997 was 497 fish/m<sup>2</sup> and 0.45 kg/Lpm. The lower number of coho reared in the control pond in 1996 compared to previous years was due to low adult returns in 1995 and a subsequent shortage in eggs/fry.

Fish in the treatment pond were allowed to feed on natural feed throughout the rearing period. Growth rate to achieve a release size of about 28 g (16 fpp) was calculated from the time of stocking to release, and the weekly total of food to achieve that growth rate was placed each week in three demand feeders located along one side of the pond. Filling the feeders was the only human contact the fish had during the 10-11 months of rearing in the pond. No sampling of fish was done until they began migrating. Fish in the control pond were fed a daily ration by hand. Growth rates in each pond were programmed to achieve a release size of 17 fpp.

Screens were lifted and fish allowed to emigrate volitionally beginning on April 14, 1997. Fish were allowed to emigrate until each pond was nearly empty.

Electronic fish counters (Northwest Marine Technology) were placed at the outlet of each rearing pond to count daily outmigration. When only a few hundred fish remained in each pond, the counters were removed and fish were forced out. Samples of these fish were also taken.

One day each week a minimum of 15 smolts from each pond were captured after passing through the fish counter. These fish were sacrificed and gill epithelial tissue was removed and handled as per methods described by Schrock et al (1994). Samples were placed in SEI medium before freezing on dry ice. Likewise, blood was collected from the same fish and centrifuged at 3000 rpm for 10 minutes to separate platelets from plasma. The plasma samples were decanted and frozen on dry ice for later analysis of plasma chloride levels. Within two hours of collecting and freezing the samples, they were express shipped to personnel of the United States Geological Survey, Columbia River Research Laboratory, Cook, WA for analysis. Analysis of the gill samples for gill Na<sup>+</sup>, K<sup>+</sup>-ATPase activities is described by Schrock et al (1994). Analysis of plasma chloride levels was done using a hand-held spectrophotometer.

Coded wire tags were applied to all 30,000 fish in the treatment pond and 30,000 of the 260,000 in the control pond. Tagging was done in May 1996 for the treatment group and the fall of 1996 for the control group. Northeast Pacific fisheries and river escapement will be sampled for the presence of marked fish and the tag recovery data reported to the Pacific State Marine Fisheries Commission, in Portland, Oregon. These data will become available for analysis from 1999-2001.

### Results

#### *Pond Construction*

The treatment pond was successfully filled with pit-run gravel over its entire length. Woody debris in the form of small alder tree tops, fir branches, and some logs were placed throughout the pond to cover approximately 25% of the total pond bottom. Branches were tied in bundles and placed against concrete bases that are used to anchor poles that support bird netting. These structures simulated log jams. Alder tops were placed around each demand feeder station to provide cover near the exogenous food source, however a small area around the feeder rods was left open to allow for sampling later in

the year if necessary. The inlet of the pond is a small stream that narrows through a concrete gate structure. This is the only area of the pond with noticeable water velocity. Brush bundles were placed in the area of flowing water as well. Bird netting covered the entire pond surface, although it was not totally effective in preventing predation from herons.

The natural rearing pond contained several types of habitat. One type consisted of an area free of LWD and consisting of coarse cobbler (pit-run gravel). A second type consisted of the gravel and LWD. In addition, cut banks were found on the lateral edge of the pond, and portions of these cut banks had LWD or macrophyte vegetation in association. Substrate types in the cut bank areas consisted of gravel or silt. At the pond outlet, the pond bottom tended to be silted in and there was less LWD located in this area.

The control pond used in 1996-97 is also the adult holding pond. Because of the dual purpose of the pond, fry were reared in concrete raceways and not stocked into the pond until early-February 1997. No modifications were made to the pond.

#### *Fish Behavior*

We observed fish behavior on two occasions by snorkeling in the NATURES pond. The first time was several hours after stocking and the second time occurred 4 months after stocking. The most significant observed behavior was: Fish after initial introduction to the pond tended to school, particularly in the upper portion of the pond, and avoid the structure. Fish seemed to be curious about the divers and would approach each diver. If a diver moved a hand or arm rapidly the fish would flee rapidly. At 4 months, the schooling behavior persisted but each school consisted of smaller numbers of individuals and some fish were found in the LWD, whereas schools were more localized around the LWD. The schools also seemed less curious of the divers and would stay at least 4-10 feet away. Divers making sudden movements elicited a more controlled fright response from the fish, in other words, the school would move away in a slower and more deliberate manner. Also at 4 months individual fish tended to be found in the upper portion of the pond, whereas schools of fish were found in the middle and lower portion. Individual fish seemed to have staked out territories and developed typical natural coho coloration, including prominent

white markings on the anterior portion of the dorsal and anal fins. However, fish that were in schools tended not to have this type of coloration. The schooling behavior in the middle and lower portions of the pond may have been due to the location of the feeders in this area, the slower water velocity, greater depth, or some unknown factors. Fish actively fed from the demand feeders, to which they had no previous exposure.

We observed the coho in the treatment pond during the summer of 1996 and the spring of 1997 from the bank of the pond only. Fish were seen actively feeding on surface insects and from the demand feeders. The hatchery crew noted large feeding activities on natural insects throughout the summer, particularly at dusk. We found it difficult to observe fish in the pond from the bank unless they were near the feeders. It is our feeling that the coloration of the fish as viewed from above, blended in well with the bottom coloration.

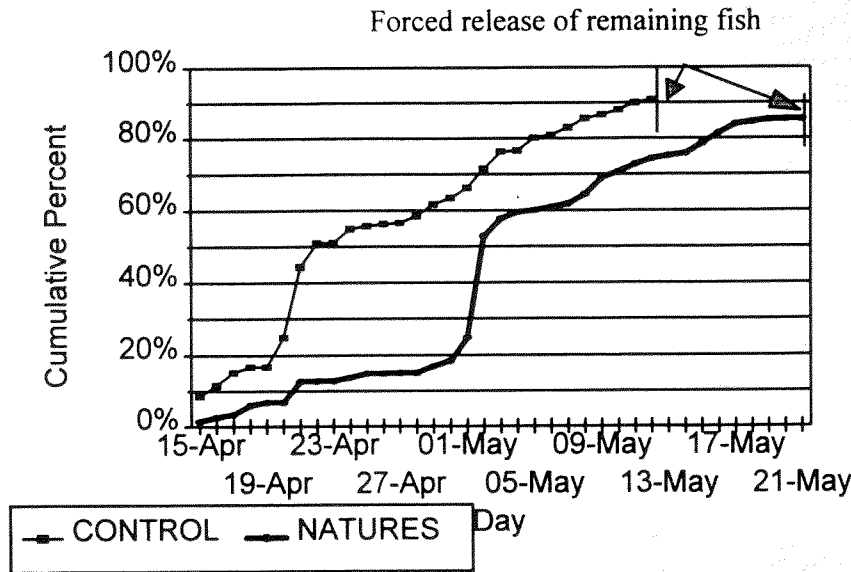
#### *Outmigration*

Electronic fish counters monitored the number of fish migrating from the ponds each day. However, when remaining fish were forced out of the ponds on May 13 (control) and May 22 (NATURES) the fish counters were not used and an estimate of the remaining population was made. Figure 1 shows the cumulative percent of the total population migrating each day. The fish in the control pond migrated at a faster rate initially than the NATURES fish. Fifty percent of the population in the control pond had migrated by 22 April in comparison to 2 May for the NATURES pond. The rate of emigration from the NATURES pond increased dramatically from May 2-5, but then maintained a steady rate thereafter, which was similar to the rate of migration from the control pond. Approximately 91% of the control pond fish and 85% of the NATURES pond fish exited volitionally.

Mean size of captured outmigrants from either the control or NATURES pond varied among weeks showing no trends in size (Table 1). Apparently, the mean size at capture reflected a threshold size at which the fish in either pond reached before migration. Likewise, condition factors of migrants from either pond showed no clear trends over time.

Because we sampled on only day each week, and because the configuration of the outlets for each pond differed, it

# Evaluation of Semi-Natural Rearing for Coho Salmon



**Figure 1.** Cumulative percent of coho population migrating by day from the control and NATURES pond. Arrows indicate day at which remaining population was forced from the rearing pond.

was not easy to assure that adequate samples were obtained. Fish from the NATURES pond were captured approximately 30 m downstream of the outlet of the pond. A seine net (0.25 in mesh) was stretched across the outlet stream on the previous evening, and fish were then sampled from a small pool between the outlet pipe and the seine net (approximately 10 feet). It is probable that this barrier net was not completely fish tight and some fish escaped capture even though the fish counter indicated adequate numbers of fish having migrated during the period. It is also possible that predators concentrated during the evening hours in the area upstream of the seine. Because the outlet of the control pond emptied directly into the river it was not possible to sample fish as they exited the outlet pipe. Instead fish were netted between the outlet screen and the outlet pipe after they had exited through the counter. On days when we were unable to obtain adequate samples from this pond it may have been due to fish not holding in this area or our inability to capture the fish.

Figure 2 shows the mean levels of ATPase activity. We were unable to obtain adequate numbers of fish from the

natures pond on April 22 and from the control pond on April 30. All fish were released from the Control pond by May 13, and from the treatment pond on May 22. In two of the three dates where we obtained adequate samples of fish from both ponds, fish originating from the natures pond had higher ATPase activities, despite being smaller sized than the control fish. Across all sample periods the fish from the natures pond had relatively similar ATPase activities with the exception of the first sample taken on April 16, which was much lower. In contrast, ATPase activities of control fish varied much more among periods. Thus, there was no apparent trends in either group relating to ATPase activities over time or size of migrants. Based on these results we assume that sampling methods had no influence on results and that we sampled actively migrating smolts each week, thus, explaining the relatively similar activities within groups and sample periods.

Coded wire tag recoveries of the first years release will occur in 1997 and 1998. We plan to release groups from the control and treatment pond for three successive years (1995-1997 broods).

Table 1. Length, weight, and condition factors for migrating fish captured from the control and NATURES pond during spring, 1997 and used in the analysis of gill Na<sup>+</sup>-K<sup>+</sup>ATPase activities on the day listed. Standard deviations are in parentheses.

CONTROL	4/16	4/22	4/30	5/7	5/13	5/22
Mean Length (SD)	140.5 (6.2)	145.5 (11.2)	152 (1 fish)	148.7 (9.2)	143.1 (14.1)	No sample
Mean Weight (SD)	28.8 (3.5)	32.5 (11.2)	39.7 (1 fish)	34.8 (6.2)	30.2 (7.8)	No sample
Mean K Factor (SD)	1.03 (0.03)	1.02 (0.05)	1.13 (1 fish)	1.05 (0.05)	1.01 (0.08)	No sample
NATURES						
Mean Length (SD)	146.5 (7.9)	No sample	141.4 (5.6)	144.9 (7.9)	142.2 (9.6)	125.6 (9.9)
Mean Weight (SD)	33.4 (5.9)	No sample	28.9 (3.7)	32.1 (5.7)	28.9 (6.0)	28.7 (6.0)
Mean K Factor (SD)	1.05 (0.03)	No sample	1.03 (0.03)	1.05 (0.04)	0.99 (0.05)	0.99 (0.05)

### Acknowledgments

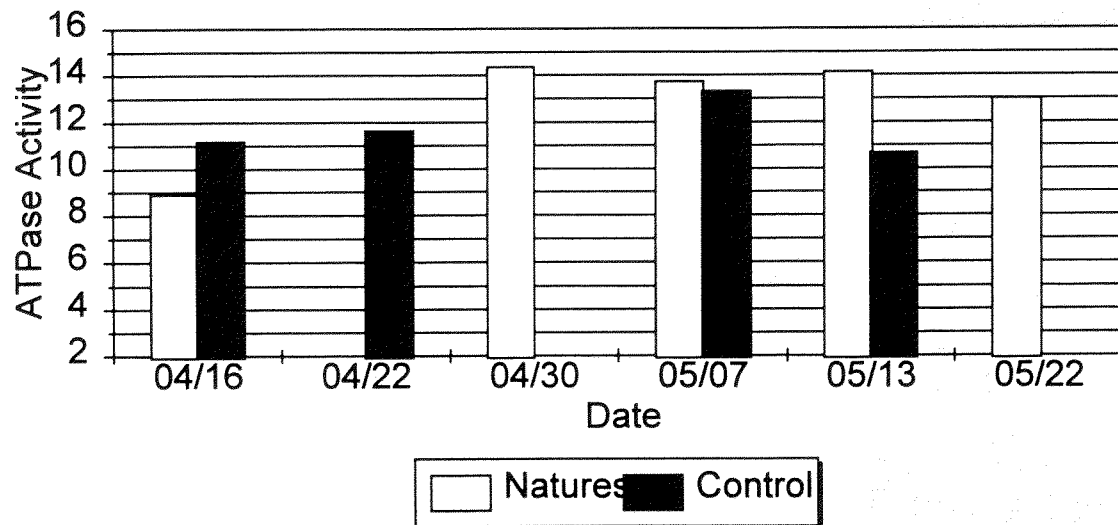
We wish to thank the National Marine Fisheries Service for providing the funding for this study. We also thank the crew of the Elochoman Hatchery for their cooperation and dedicated efforts to make this study flow smoothly.

### Literature Cited

- Brauner, C.J., G.K. Iwama, and D.J. Randall. 1994. The effect of short-duration seawater exposure on the swimming performance of wild and hatchery-reared juvenile coho salmon (*Oncorhynchus kisutch*) during smoltification. *Canadian Journal Fishery and Aquatic Sciences*, 51:2188-2194.
- Maynard, D.J., T.A. Flagg, and C... Mahnken. 1996. Development of a natural rearing system to improve supplemental fish quality, 1991-1995. Progress Report, U.S. Department of Energy, Bonneville Power Administration, Portland, Oregon. 216 pp.
- Shrimpton, J.M. N.J. Bernier, G.K. Iwama, and D.J. Randall. 1994. Differences in measurements of smolt development between wild and hatchery-reared juvenile coho salmon (*Oncorhynchus kisutch*) before and after saltwater exposure. *Canadian Journal Fishery and Aquatic Sciences*, 51:2170-2178.



## ATPase Levels 1997 Samples



**Figure 2.** Mean  $\text{Na}^+\text{-K}^+$  ATPase activities of coho from the natures and control ponds captured during outmigration on various dates in 1997.



# Evaluating the Effects of Complex Rearing Habitats on Juvenile Fall Chinook The Bingham Creek Experience

Steve Schroder<sup>1</sup> and C.M. Knudsen  
*Washington Department of Fish and Wildlife*

B.A. Berejikian and E.P. Tezak  
*National Marine Fisheries Service*

**Abstract**— Previous studies have showed that the post-release survival of cultured fall chinook can be improved when fish are reared in raceways containing gravel substrates, intra-gravel filtration, underwater feeders, in-water structure, and overhead cover. Our objectives were to determine whether similar gains could be achieved by using only a few of these components or by reducing the length of time the fish were cultured in such environments.

In 1996, we evaluated the effects of seven different rearing treatments on the in-culture performance and post-release survival of fall chinook. In the control or OCT (Optimal Conventional Treatment) case, the fish were fed by hand from the surface and nothing was added to the two meter circular tanks we used as rearing vessels. The remaining six treatments all had 90 percent of their surface area covered with camouflage netting. In one treatment, this was the only added feature. An additional single element, either an underwater feeder, a filter that was covered with a scattered layer of pea gravel, or two submerged panels of camouflage netting were added to create three additional rearing regimes. The remaining two treatments possessed covers, underwater feeders, intra-gravel filters, and in-water structure, however fish were reared under these conditions for varying periods of time. Those experiencing a full NATS (Natural Rearing Systems) treatment were held throughout their entire 92-105 day rearing period. Fish exposed to the LNAT (Limited) treatment, on the other hand, were reared under OCT conditions and then exposed to a NAT regime over the last 30 days of their rearing period. None of the rearing treatments appeared to affect the growth of the fish.

Differences, however, did occur in their in-culture survival. Fish reared in tanks without substrates or in water structure had mortality rates that were less than 1 percent, fish held in tanks with in-water structure experienced slightly higher mortalities (1.5%) while the NAT, LNAT, and Cover-substrate tanks had the highest mortality rates (up to 20%). The rearing environments also affected the color patterns of the fish. Individuals from the NAT and LNAT tanks had pronounced parr marks, colored fins, and heavy melanic spotting and thus appeared to be more cryptic than fish produced from the other rearing treatments. The post-release survival of fish originating from each treatment was evaluated by making two separate releases into Bingham Creek and allowing the fish to migrate 21 km before being recaptured. In the first release, fish reared under the NATS regime had a 16% higher survival rate than those held in the OCT tanks. No other survival differences were seen. In the second release, fish originating from the LNAT tanks had significantly lower survival rates than those originating from the other rearing treatments. Just prior to being released, these fish had experienced a *Costia* outbreak and we speculate that this may have adversely affected their survival. In 1997, we examined the effects of four different substrates and a NATS treatment on the in-culture performance and post-release survival of fall chinook at Bingham Creek. As in 1996, the fish were liberated into Bingham Creek after a 3-month rearing period.

The in-stream survival rates of these groups have not yet been evaluated. Distinct differences in the color patterns of the fish were noted and mortality was slightly higher than two percent across all the groups. We did see slightly more fin and opercle erosion on fish reared in the NATS tanks, however.

---

<sup>1</sup> Steve elected to only have the abstract of his talk published in the Proceedings.



# Seminatural Raceway Habitat Increases Chinook Salmon Post-release Survival

Desmond J. Maynard, Eugene P. Tezak, Michael Crewson,  
Deborah A. Frost, Thomas A. Flagg

*Resource Enhancement and Utilization Technology Division, Northwest Fisheries Science Center  
National Marine Fisheries Service, National Oceanic and Atmospheric Administration  
2725 Montlake Boulevard East, Seattle, Washington 98112*

Steve L. Schroder, Chuck Johnson  
*Washington State Department of Fish and Wildlife  
600 Capitol Way North, Olympia, Washington 98501-1091*

Conrad V. W. Mahnken  
*Resource Enhancement and Utilization Technology Division, Northwest Fisheries Science Center  
National Marine Fisheries Service, National Oceanic and Atmospheric Administration  
2725 Montlake Boulevard East Seattle, Washington 98112*

**Abstract** — There is growing concern that hatchery-reared salmonids die at higher rates than their wild-reared counterparts. We hypothesize that seminatural raceway habitats (raceways fitted with overhead cover, instream structure, and substrate) will improve the Post-release survival of hatchery-reared salmonids by better acclimating fish to their Post-release environment. Three experiments were conducted to evaluate if seminatural raceway habitats increased the Post-release survival of chinook salmon (*Oncorhynchus tshawytscha*).

In the first experiment, ocean type chinook salmon were reared from swim-up to smoltification (4 months) in 400-L raceways with opaque overhead cover, plastic aquarium plant structure, and sand or gravel substrates. During culture, the seminaturally-reared fish exhibited escalated agonistic behavior compared to conventionally-reared fish. At the end of rearing, the skin hue and chroma of seminaturally-reared fish differed significantly ( $P < 0.05$ ) from conventionally-reared fish. This effectively enhanced their ability to camouflage to their post-release stream background. When released into a small coastal stream (Little Anderson Creek on Hood Canal) the survival to a collection weir 2.2 km downstream was significantly ( $P < 0.05$ ) higher (51%) for seminaturally-reared than conventionally-reared fish.

In the second experiment, an acclimation approach to seminatural raceway habitat rearing was evaluated. Stream type chinook salmon were initially reared in barren circular tanks for 9 months after swimup. These fish were then transferred to either 400-L seminatural or conventional raceway tanks for the final 4 months of experimental rearing. The seminatural habitat in this experiment consisted of opaque overhead covers, ornamental junipers for structure, and gravel substrate. When released as smolts into the Yakima River under clear water conditions, the Post-release survival of seminaturally-reared fish to a collection weir 225 km downstream was significantly higher (24%;  $P < 0.05$ ) than conventionally-reared fish. When released under turbid water conditions, there was no significant difference in the Post-release survival of the two treatment groups.

In the final experiment, culture vessel size was increased to 5,947-L, and ocean type chinook salmon were reared for about 3 months (from swimup to smoltification) in raceways outfitted with camouflage net covers, fir tree structure, gravel substrate, and an underwater feed-delivery system. At the end of rearing, the skin coloration of seminaturally-reared fish again appeared to be better suited for blending into the natural stream background. When released into a tributary of the Satsop River (Bingham Creek), the seminaturally-reared fish averaged significantly higher (26%;  $P < 0.05$ ) Post-release survival to a collection weir 21 km downstream than their conventionally-reared counterparts.

In all three experiments, seminaturally-reared fish developed more natural camouflage coloration than conventionally-reared fish. We hypothesize that the higher Post-release survival of seminaturally-reared fish resulted from their lower predator vulnerability due to their enhanced crypsis in the stream environment. Seminatural raceway habitats provide fish culturists a tool to increase the Post-release survival of salmon released in fisheries enhancement and conservation programs.

## Introduction

NATURES (Natural Rearing Enhancement System) studies developed by National Marine Fisheries Service and Washington Department of Fish and Wildlife scientists are aimed at developing salmon culture techniques that rear fish

in a more natural environment. Traditionally, salmon are reared in barren concrete raceways that lack natural substrate, instream structure, or overhead cover. The fish are fed in an unnatural manner with artificial feeds that are mechanically or hand broadcast across the water surface. This traditional approach can markedly increase the egg-to-

smolt survival of hatchery-reared fish over that of wild-reared salmon. However, once hatchery-reared fish are released into the wild, their smolt-to-adult survival is usually much lower than wild-reared salmon. In the best fall chinook salmon hatchery programs, only 0.1- 5.0 % of the released fish survive (note: Survival = catch + escapement using CWT) (Mahnken et al 1997).

The lower Post-release survival of hatchery-reared fish may stem from how their behavior and morphology differs from wild-reared salmon. After release, hatchery-reared fish are inefficient foragers and are often found with empty stomachs or stomachs that are filled with indigestible debris (Miller 1953, Hochachka 1961, Reimers 1963, Sosiak et al. 1979, Myers 1980, O'Grady 1983, Johnsen and Ugedal 1986). Their social behavior also differs, with hatchery-reared fish congregating at higher densities, being more aggressive, and displaying less territory fidelity than wild-reared fish (Fenderson et al. 1968, Bachman 1984, Swain and Riddle 1990). In the natural environment, this results in hatchery-reared fish spending more time in high risk aggressive behavior and less time in beneficial foraging behavior than their wild-reared counterparts. Hatchery-reared fish are also more surface oriented than wild-reared salmonids (Mason et al. 1967, Sosiak 1978). This may increase their risk of being attacked by avian predators, such as kingfishers, that search for fish near the surface. Although some of the differences observed between wild and hatchery-reared fish are innate (Reisenbichler and McIntyre 1977, Swain and Riddle 1990), many are conditioned and can be modified by altering the hatchery rearing environment.

Rearing salmon in a more stream-like environment may be a way to produce fish with more natural behavior and morphology that increases their Post-release survival. NATURES researchers have developed a stream-like rearing habitat concept that can be retrofitted to existing hatchery raceways. In this seminatural rearing habitat, the raceway bottom is covered with sand or gravel substrates matching the substrate color of streams into which the fish will be released. An under-gravel filtration system may be placed beneath the gravel substrate to assist in the biological decomposition of organic particles that become lodged in the substrate. Artificial aquatic plants or heavily branched conifers are placed throughout the raceway to create instream structure. Opaque black covers or military camouflage netting is hung 30 cm or less from the water surface to simulate the overhanging vegetation and undercut banks found along salmon stream margins.

Rearing salmon in these more stream-like habitats should prepare them for life in the natural environment they will be released into as smolts. In addition, seminatural rearing may slow down the genetic divergence (domestication) occurring between hatchery fish and the wild populations from which they were sourced. We have conducted several NATURES experiments to demonstrate that rearing salmon in seminatural raceway habitat can increase salmonid Post-release survival.

### 1992 Little Anderson Creek Release Experiment

The benefit of rearing chinook salmon in seminatural raceway habitat was first demonstrated in an experiment conducted in 1992 (Maynard et al. 1996 a, b). In this experiment, 12 400-L rectangular tanks with a grey background were each stocked with 40 fall chinook salmon swimup fry. Four of the tanks served as experimental controls. To simulate conventional raceway environments, these control tanks had grey bottoms and sides, clear plexiglass tops that provided no shade, and no instream structure except for the aeration system used to compensate for the air-driven under-gravel filtration system in the seminatural habitat tanks. In addition to the control features, four of the seminatural habitat tanks were fitted with a sand substrate on the bottom, artificial aquatic plants for instream structure, and opaque black covers to shade the fish. The other four seminatural tanks were similar, except that the sand was replaced with a pea gravel substrate spread over an under-gravel filter plate.

The fish in both types of seminaturally-reared tanks exhibited significantly ( $P = 0.046$ ) more aggressive acts (contact nips, threat nips, and chases) to one another than conventionally-reared fish (Fig. 1). This escalated aggressive activity may have been the product of the plastic aquarium plants providing more focal points around which fish could establish territories. The increased number of territorial individuals in the seminatural tanks in turn raised the number of territorial disputes that resulted in aggressive actions. The complex habitat structure of seminatural tanks thus seemed to produce aggressive activity that was more natural than that found in the conventional artificial rearing environment.

Fish in conventional tanks struck at drifting material significantly ( $P = 0.004$ ) more often than fish in seminatural tanks (Fig. 2). As the fish were not fed prior to or during the observation period, it appears this striking activity was directed at decaying food and fecal material drifting in the water column. The plants, substrates, or interstitial

## Seminatural Raceway Habitat

microorganisms in seminatural tanks seemed to provide the fish with a more sanitary rearing environment by removing these organic particles from the water column. The reduced level of visible drifting debris in the seminatural tanks seems to be responsible for the decreased striking activity observed in these tanks.

The external body coloration of conventionally- and seminaturally-reared fish strongly differed (Fig. 3). Conventionally-reared fish had a uniform light tan coloration that blended in with the homogenous light grey background of their rearing tanks. In contrast, seminaturally-reared fish had a dark brown skin coloration that matched the sand and gravel substrates they were reared over. The parr marks and dorsal spotting were also more pronounced in seminaturally- than conventionally-reared fish. In a stream or river environment these features would help the fish visually blend into the background, with the parr marks breaking up the overall body outline and dorsal spotting mimicking small dark stones randomly scattered over the bottom. After release, visually hunting predators should have more difficulty detecting seminaturally- than conventionally-reared salmon, because the former has developed camouflage that blends in with the stream background.

After the fish were reared from the swimup fry to smolt stage, they were tagged and released into Little Anderson Creek to determine if seminatural rearing enhanced Post-release survival. In the 2.2 km migration corridor from the release site to the weir, the survival of seminaturally-reared fish was 51% higher than conventionally-reared fish (Fig. 4a). This survival difference was most likely the result of seminaturally-reared fish having better camouflage coloration in the stream environment than conventionally-reared fish.

### 1994 Yakima River Release Experiment

In 1994 a second experiment was conducted to determine if the benefits of seminatural rearing could be obtained when fish were conditioned only during the last part of the freshwater rearing cycle (Maynard et al. 1996 a, c). In this experiment, each 400-L rearing tank was stocked with 80 spring chinook salmon fry that had been previously reared for more than 8 months in uniform blue colored circular tanks. Twelve of the tanks were conventional raceway habitats with grey backgrounds, clear covers, and no instream structure. The other 12 tanks were seminatural raceway habitats with pea gravel substrates, under-gravel

filter plates, opaque overhead cover, and juniper trees for instream structure. After 4 months of experimental rearing, the fish were tagged and released as smolts into the Yakima River to evaluate their Post-release survival.

The fish were released into the Yakima River under two distinctly different water conditions. In the first release, the Yakima River water was running clear, and visually hunting predators would have to detect the fish against the heterogenous background coloration of the river bottom. Under these conditions, fish reared in seminatural tanks would theoretically have the best camouflage coloration. The second release occurred when the water in the Yakima River was very turbid. This condition produced a bright uniform background coloration against which visual hunting predators would have to detect their prey. Theoretically, conventionally-reared fish should have the camouflage advantage, with their light uniform coloration blending in better with the turbid water background. The darker seminaturally-reared fish should be slightly easier to detect against this bright uniform background. Each release was paired, with all the fish from six seminatural and six conventional rearing tanks being released on each day.

When released into the Yakima River, seminaturally- and conventionally-reared fish were observed to maintain their color differences, even after several hours of being held in a common transport tank. As theoretically expected in the clear water release, seminaturally-reared fish had a significantly higher (24%;  $P = 0.036$ ) survival than conventionally-reared fish (Figure 4b). Under turbid water conditions, conventionally-reared fish had a slightly (10%), but statistically insignificant ( $P > 0.05$ ), higher recovery rate than seminaturally-reared fish. Fish from both rearing treatments migrated down river at similar speeds, although the downstream migration of fish was faster under turbid than clear water conditions. These findings indicate that an acclimation approach, in which fish are held in seminatural raceway habitats for only the last few months before release, can be successfully used to increase chinook salmon Post-release survival.

### 1994 Bingham Creek Release Experiment

Another experiment was initiated in 1994 to determine if the seminatural raceway habitat concept could be scaled up to production size raceways (Maynard et al. 1996 a, 1996,d). This experiment was conducted in six fiberglass raceways that were 6.4-m long by 1.5-m wide by 0.6-m deep at the Washington Department of Fish and Wildlife's (WDFW)

Bingham Creek Hatchery. In February 1994, each raceway was stocked with 6,000 fall chinook salmon swimup fry. The three conventional raceways were again barren grey tanks containing only water and fish. The seminatural raceway habitats had pea gravel substrate that covered under-gravel filter plates and heavily branched conifers for instream structure. The conventional raceways were only covered with a translucent bird net that prevented predation, while a shade-producing military camouflage net covered 80% of the seminatural raceway surface. In this experiment, an underwater feeder was added to the seminatural raceways. The fish in the conventional raceways were handled from the surface as in the previous experiments. The fish were reared in these environments until June 1994. The fish were tagged and three paired releases at a point 21 km above a collection weir were made into Bingham Creek.

As in the previous experiment, seminaturally-reared fish developed camouflage coloration better suited to the clear water stream environment than conventionally-reared fish (Figure 5). Once again, the fish maintained these color differences even after being held in a transport tank for several hours prior to release. During the first week after release all fish intercepted at the weir had either conventionally- or seminaturally-reared color patterns. Fish with intermediate color patterns were not seen until 2 weeks after release. This suggests conventionally-reared fish could not develop proper camouflage coloration suitable for the natural stream environment for at least a week after release.

In the three Bingham Creek releases, seminaturally-reared fish had 13.5 to 66.5% higher Post-release survivals than conventionally-reared fish. On average the survival of seminaturally-reared fish was 26% higher than conventionally-reared fish (Fig. 4c). These findings suggest that the seminatural rearing habitat concept developed in 400-L raceways can be successfully scaled up and still increase Post-release survival.

### Conclusion

These three experiments demonstrate that rearing salmonids in seminatural habitat with overhead cover, instream structure, and natural substrate markedly increases hatchery-reared salmonid Post-release survival. This seminatural habitat is slightly harder to maintain than conventional tanks, with overhead covers and instream structure needing to be removed during raceway cleaning and fish handling operations. As in conventional tanks, gravel substrates in seminatural tanks are cleaned by vacuuming, but require

somewhat longer to clean as the gravel must be separated from decaying food and feces during this process. The Post-release survival benefits of seminatural rearing far outweigh these increased maintenance requirements.

The Post-release survival advantage offered by seminatural rearing has numerous benefits (Maynard et al. 1995). Increased survival can reduce the number of broodstock that startup and supplementation hatchery programs must remove from the natural spawning population. Operational costs can be reduced by decreasing the number of smolts a facility must produce to yield a given number of recruits to the fishery. Releasing fewer smolts can reduce Post-release competition between wild and hatchery-reared salmon. Finally, the higher Post-release survival of seminaturally-reared fish should produce more recruits to the fishery and spawning population. This increased production can then be used by managers to increase harvest, meet mitigation goals, or speed the rebuilding of endangered and threatened salmon runs.

### Acknowledgments

These studies were supported in part by the Bonneville Power Administration through interagency agreement with the National Oceanic and Atmospheric Administration. We thank James Hackett and Richard Kerr for constructing the various NATURES fish culture systems. We thank Gail McDowell for reviewing the manuscript and improving its readability. Finally we would extend our greatest appreciation to Michael Kellett, Glen Snell, Gail McDowell, Lee Harrell, Carlin McCauley, Michael Wastel, James Hackett, Curt Knudsen, Robert Iwamoto, Donald VanDornick, and David Kuligowski for assisting us in sampling and tagging fish over the course of these experiments.

### References

- Bachman, R. A. 1984. Foraging behavior of free-ranging wild and hatchery brown trout in a stream. *Trans. Am. Fish. Soc.* 113:1-32.
- Fenderson, O. C., W. H. Everhart, and K. M. Muth. 1968. Comparative agonistic and feeding behavior of hatchery-reared and wild salmon in aquaria. *J. Fish. Res. Board Can.* 25:1-14.

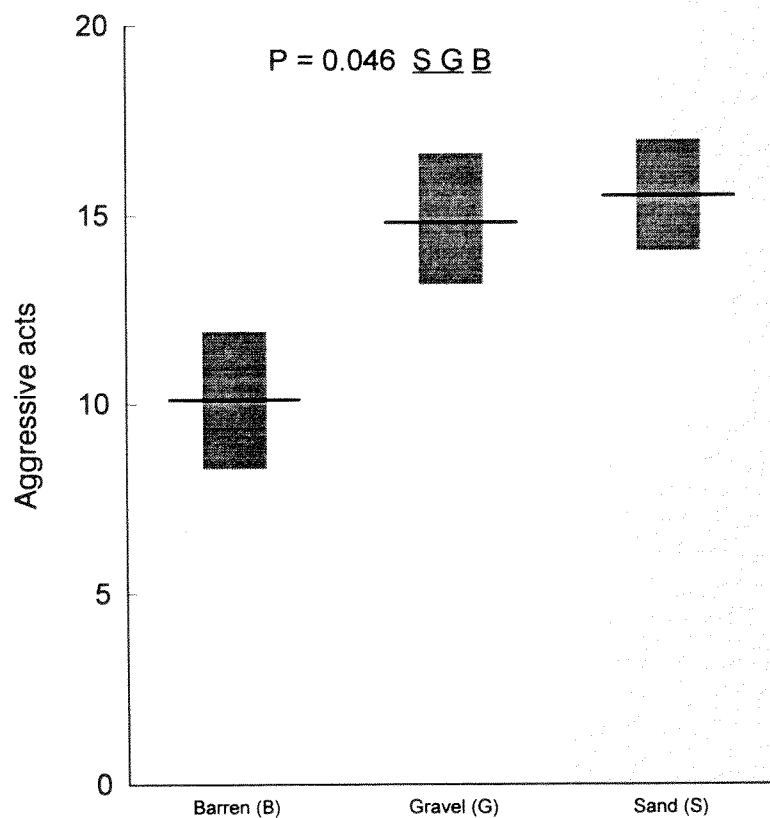


# Seminatural Raceway Habitat

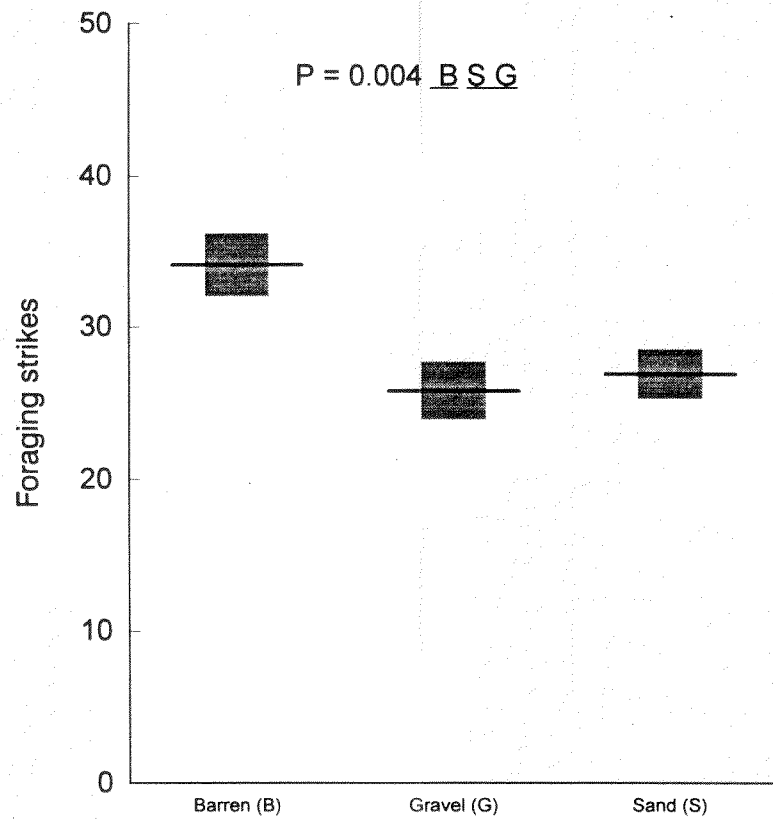
- Johnson, B. O., and O. Ugedal. 1986. Feeding by hatchery-reared and wild brown trout, *Salmo trutta* L., in a Norwegian stream. *Aquacult. and Fish. Manage.* 20:97-104.
- Hochachka, P. W. 1961. Liver glycogen reserves of interacting resident and introduced trout populations. *J. Fish. Res. Board Can.* 18:125-135.
- Mahnken, C., G. Ruggerone, W. Waknitz, and T. Flagg. 1997. A historical perspective on salmonid production from Pacific rim hatcheries. *N. Pac. Anadr. Fish. Comm. Bull.* 1:38-53.
- Mason, J. W., O. M. Brynildson, and P.E. Degurse. 1967. Comparative survival of wild and domestic strains of brook trout in streams. *Trans. Am. Fish. Soc.* 9:313-319.
- Maynard, D. J., T. A. Flagg, and C. V. W. Mahnken. 1995. A review of seminatural culture strategies for enhancing the postrelease survival of anadromous salmonids. *Am. Fish. Soc. Symp.* 15:307-314.
- Maynard, D. J., T. A. Flagg, C. V. W. Mahnken, and S. L. Schroder. 1996a. Natural rearing technologies for increasing postrelease survival of hatchery-reared salmon. *Bull. of Natl. Res. Inst. of Aquacult., Suppl.* 2:71-77.
- Maynard, D. J., M. S. Kellet, D. A. Frost, E. P. Tezak, W. C. McCauley, T. A. Flagg, and C. V. W. Mahnken. 1996b. The behavior and postrelease survival of fall chinook salmon reared in conventional and seminatural raceways, 1992. Pages 21-28 in D. J. Maynard, T. A. Flagg, and C. V. W. Mahnken, editors. Development of a natural rearing system to improve supplemental fish quality, 1991-1995 progress report to the Bonneville Power Administration, Contract DE-A179-91-BP20651, 216 p. (Available from Bonneville Power Administration, P.O. Box 3621, Portland, OR 97208).
- Maynard, D. J., M. Crewson, E. P. Tezak, W. C. McCauley, and T. A. Flagg. 1996c. The postrelease survival of Yakima River spring chinook salmon acclimated in conventional and seminatural raceways, 1994. Pages 66-77 in D. J. Maynard, T. A. Flagg, and C. V. W. Mahnken, editors. Development of a natural rearing system to improve supplemental fish quality, 1991-1995 progress report to the Bonneville Power Administration, Contract DE-A179-91-BP20651, 216 pages (Available from Bonneville Power Administration, P.O. Box 3621, Portland, OR 97208).
- Maynard, D. J., M. Crewson, E. P. Tezak, W. C. McCauley, S. L. Schroder, C. Knudsen, T. A. Flagg, and C. V. W. Mahnken. 1996d. The postrelease survival of Satsop River fall chinook salmon reared in conventional and seminatural raceway habitats, 1994. Pages 78-97 in D. J. Maynard, T. A. Flagg, and C. V. W. Mahnken, editors. Development of a natural rearing system to improve supplemental fish quality 1991-1995 progress report to the Bonneville Power Administration, Contract DE-A179-91-BP20651, 216 pages. (Available from Bonneville Power Administration, P.O. Box 3621, Portland, OR 97208).
- Miller, R. B. 1953. Comparative survival of wild and hatchery-reared cutthroat trout in a stream. *Trans. Am. Fish. Soc.* 83:120-130.
- Myers, K. 1980. An investigation of the utilization of four study areas in Yaquina Bay, Oregon, by hatchery and wild juvenile salmonids. M.S. Thesis, Oregon State University, Corvallis, 233 pages.
- O'Grady, M. F. 1983. Observations on the dietary habits of wild and stocked brown trout, *Salmo trutta* L. in Irish lakes. *J. Fish Biol.* 22:593-601.
- Reimers, N. 1963. Body condition, water temperature, and over-winter survival of hatchery reared trout in Convict Creek, California. *Trans. Am. Fish. Soc.* 92:39-46.
- Reisenbichler, R. R., and J. D. McNytire. 1977. Genetic differences in growth and survival of juvenile hatchery and wild steelhead trout, *Salmo gairdneri*. *J. Fish. Res. Board Can.* 34:123-128.
- Sosiak, A. J. 1978. The comparative behavior of wild and hatchery-reared juvenile Atlantic salmon (*Salmo salar* L.). M.S. Thesis, University of New Brunswick, Fredrickton, 198 pages.

- Sosiak, A. J., R. G. Randall, and J. A. McKenzie. 1979. Feeding by hatchery-reared and wild Atlantic salmon (*Salmo salar*) parr in streams. J. Fish. Res. Board Can. 36:1408-1412.
- Swain, D. P., and B. E. Riddell. 1990. Variation in agonistic behavior between newly emerged juveniles from hatchery and wild populations of coho salmon, *Oncorhynchus kisutch*. Can. J. Fish. and Aquat. Sci. 47:566-571.

# Seminatural Raceway Habitat

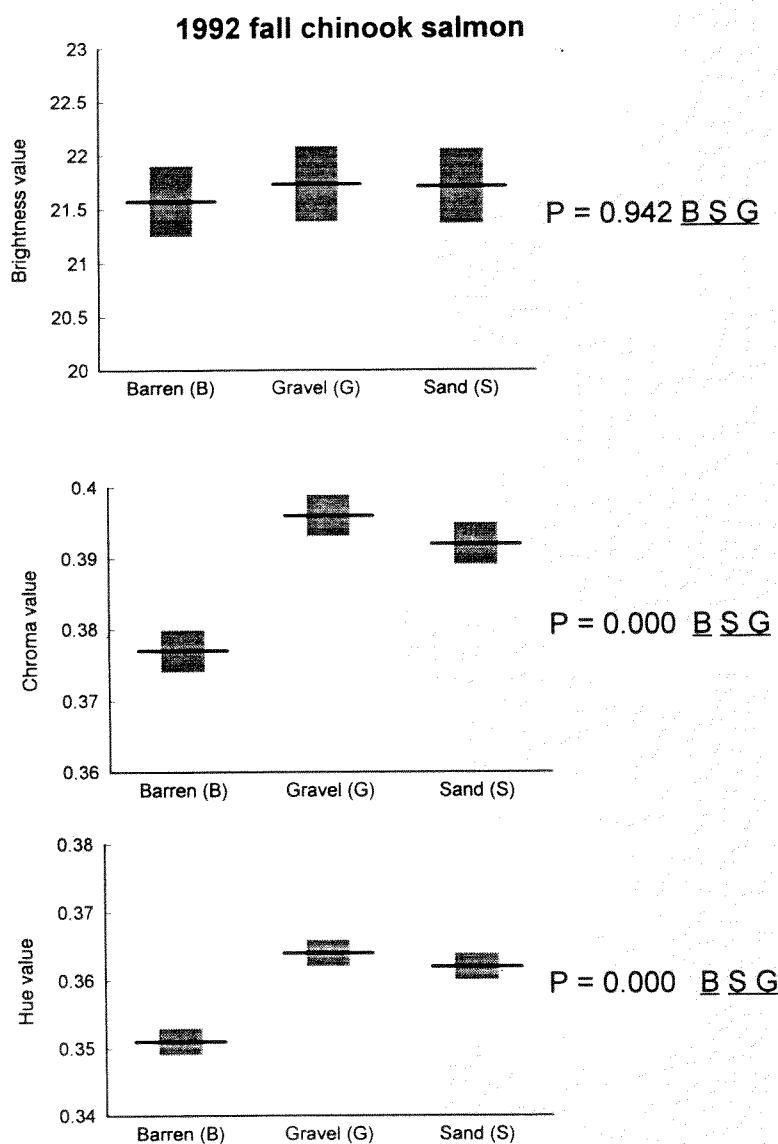


**Figure 1.** Mean number of aggressive acts per 10 minute scan sample in conventional and seminatural raceway habitat tanks in 1992 Little Anderson Creek release experiment. (Bars are mean values and boxes are standard error.)

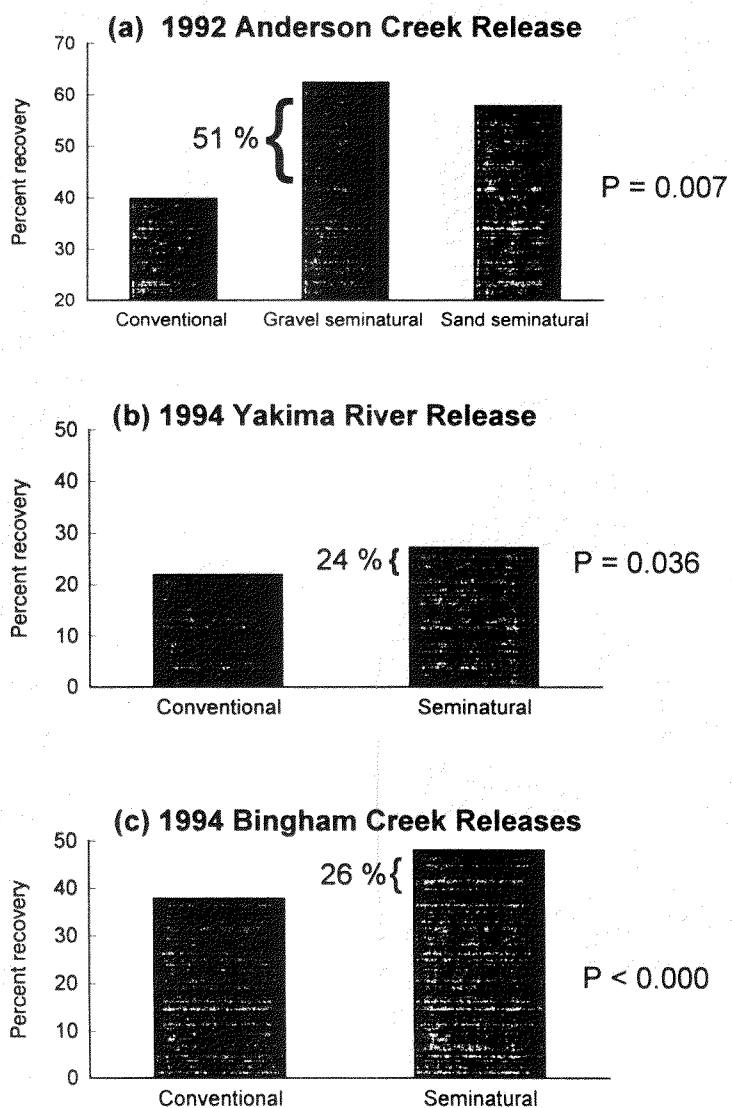


**Figure 2.** Mean number of particle strikes per 10 minute scan sample in conventional and seminatural raceway habitat tanks in 1992 Little Anderson Creek release experiment. (Bars are mean values and boxes are standard error.)

Seminatural Raceway Habitat

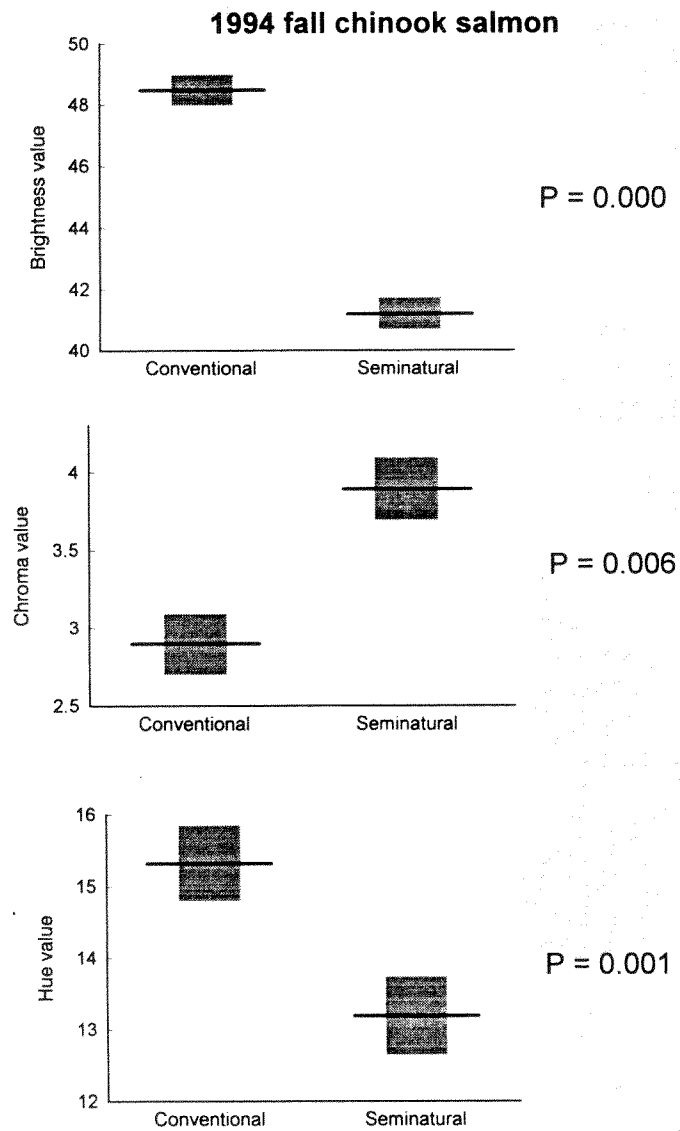


**Figure 3.** Mean brightness, chroma, and hue value for ocean type chinook salmon reared in conventional and seminatural raceway habitats in 1992 Little Anderson Creek release experiment. (Bars are mean values and boxes are standard error.)



**Figure 4.** Percent recovery of conventionally- and seminaturally-reared chinook salmon from clearwater releases into: (a) Little Anderson Creek in 1992; (b) the Yakima River in 1994; and (c) Bingham Creek in 1994.

Seminatural Raceway Habitat



**Figure 5.** Mean brightness, chroma, and hue value for ocean type chinook salmon reared in conventional and seminatural raceway habitats in 1994 Bingham Creek release experiment. Bars are mean values and boxes are standard error.)





# Session IV

## **Pathology/Disease**

Session Chair:

Dr. Richard Holt  
(Oregon Department of  
Fish and Wildlife)

# Results from a Chloramine-T Clinical Efficacy Trial To Control Mortality Among Fall Chum Salmon Caused by Bacterial Gill Disease

James D. Bowker

U.S. Fish & Wildlife Service, Bozeman Fish Technology Center, National INAD Office,  
4050 Bridger Canyon Rd., Bozeman, Montana 59715

Larry Telles

U.S. Fish & Wildlife Service, Quilcene National Fish Hatchery  
281 Fish Hatchery Rd., Quilcene, Washington 98376

**Abstract** — A clinical field trial was conducted at the Quilcene National Fish Hatchery (Quilcene, Washington) to evaluate the efficacy of chloramine-T to control mortality among fall chum salmon (*Oncorhynchus keta*) fingerlings infected with bacterial gill disease (BGD). A total of six test units holding approximately 14,000 - 17,000 fish per unit were used during the study. Three test units were treated a total of three times each with 12 mg/L chloramine-T for 60 minutes using a standing bath system on alternate days. The remaining three units served as untreated controls. Fish used in the study were diagnosed with a moderate to severe case of BGD based on results from microscopic examination of stained gill squash samples. The causative agent was a gram-negative long, filamentous bacteria. Mean total mortality at the end of the 19-d study was significantly lower ( $<0.05$ ) among treated units (1,298; 8.6%) than among non-treated units (16,014; 97.6%). The chloramine-T treatment regime used clearly was efficacious in controlling mortality caused by BGD in fall chum salmon fingerling.

## Introduction

Bacterial gill disease (BGD) is one of the most common diseases of hatchery reared salmonids (Bullock 1990; Warren 1991) and causes more fish losses than any other bacterial disease (Bills 1988). In Ontario, this disease accounts for about 21% of all diagnostic submissions from fish farms to the Fish Pathology Laboratory of the Ontario Veterinary College (Ferguson 1991). Although death is generally from a massive infection of the gills, stressors associated with intense fish culture may predispose fish to infection. *Flavobacterium branchiophilum* is the bacteria causing most outbreaks of BGD (Wakabayashi, H, et al., 1989; Ferguson et al., 1991), however other gram-negative bacteria have also been implicated. Proliferation of gill epithelial tissue, and later the loss of gill surface by clubbing and fusing of lamellae (Bullock 1990) are often associated with this bacterial infection. The disease is characterized by acute onset, flared opercula, increased branchial rate, decreased fright response, reduction in feed uptake, and high mortality rate (Lumsden 1994). If BGD is not diagnosed and treated early, thousands of fish may die within a 24-h period (Bullock 1991).

Historically, several chemicals, including benzalkonium chlorides, available as Hyamine 1622 and 3500, diquat, and chloramine-T (Bullock 1990) have been used to try to control mortality caused by BGD. However, none of these chemicals are approved by the U.S. Food and Drug

Administration (FDA) to control mortality in freshwater fish caused by BGD. Because chloramine-T seems to be the most effective therapeutant when salmonids have BGD (From 1980; Bullock et al. 1991) it has become one of the prime candidates for approval with the FDA as a bath treatment. Currently, all use of chloramine-T in the United States is granted under a compassionate Investigational New Animal Drug (INAD) exemption. Requirements for approval of an INAD compound include, among other things, data from clinical field efficacy (pivotal) trials. Data compiled by the U.S. Fish & Wildlife Service (Bowker and Erdahl 1997) show that 10 or 15 mg/L chloramine-T administered for 60 minutes using a flow through or standing bath method on three alternate days to be a highly effective treatment regime ( $>87\%$  of the trials appeared effective). However, these data are not considered to be of sufficient quality (pivotal) to satisfy FDA/Center for Veterinary Medicine's (CVM) minimum requirement for clinical field efficacy data, a necessary component of a New Animal Drug Application (NADA) packet. Therefore, a study was designed to test the efficacy of chloramine-T to control mortality in fall chum salmon fingerlings caused by BGD.

## Methods

### Study Location and Schedule

This trial was conducted at the Quilcene National Fish Hatchery (281 Fish Hatchery Rd, Quilcene, Washington). Fish were hatched at the facility and reared in their

respective rearing units prior to initiation of the study. The trial was initiated on May 19, 1997. The treatment period covered a period extending from May 19 - 23. The post-treatment period extended from May 20 - June 6, 1997. Duration of the entire study was 19 days.

### Test Fish

Test fish were fall chum salmon from the same production lot spawned from returning adults. Fall chum salmon were spawned by hand at the hatchery on December 2, 1996. Eggs were fertilized, disinfected and incubated in trough incubators for 55 days. Eggs were then shocked, sorted and counted using a McCleary mechanical counter. A total of 5,000 eggs were placed in Heath incubator trays over a period extending from January 27 - 29, 1997. Eggs hatched on/around February 25, 1997. On April 24, 1997 fry from 24 Heath incubator trays were distributed equally among 6 test units (eggs from 4 trays per test unit). At the time fish were transferred from incubator trays to test units, average fish weight was approximately 0.37 g.

### Experimental Design

The study procedure that was followed is described in detail in a recently accepted chloramine-T pivotal field efficacy trial protocol. The study design consisted of two treatments: a single treated group and a single untreated group. Each treatment group consisted of three replicates. Fish used in both treated and untreated test units came from the same lot of fall chum salmon. The experimental design used was completely randomized. Treated fish received chloramine-T at a target dosage of 12 mg/L chloramine-T for 1-h on three alternate days using a standing bath treatment. Chloramine-T used in the study was supplied by Akzo Chemical, Inc (Chicago, Illinois). Fish in the remaining three test units received no therapeutant, but rather a sham water treatment. Mortality data were collected during the 5-d treatment and 14-d post-treatment periods. Study test units used were rectangular fiberglass troughs (4.98 m x 0.92 m x 0.61 m). Density index values were calculated using test unit dimensions with tail-screen in place, and total area was determined to be approximately 2.5 cubic meters (88.4 cubic feet). The amount of chloramine-T needed to produce desired concentrations was based on the entire tank water volume. Approximately 48,204 and 48,217 fish were randomly split among treated and untreated test units, respectively. The number of fish in each test unit,

test unit treatment condition and select culture conditions used in the study are summarized in Table 1.

Table 1. Treatment condition assigned to each test unit, number of fish/test unit, flow index and density index at study start.

Tank No.	Treatment Condition	No. Fish per Tank	Flow Index	Density Index
#2	Treated	15,261	1.12	0.19
#3	Control	17,971	1.32	0.22
#4	Control	17,667	1.30	0.22
#13	Control	13,634	1.00	0.17
#14	Treated	14,604	1.07	0.18
#15	Treated	19,252	1.41	0.24

Water flow was set and maintained at 57 L/min and the water turnover rate was determined to be 1.2 exchanges per hour. Water hardness and pH were 38 mg/L (as CaCO<sub>3</sub>) and 7.2, respectively at the start and end of the study.

### Study Procedure

Fish were reared in 6 identical tanks, under similar environmental conditions. Fish began to display behavior characteristic of BGD approximately one week prior to the start of the study, and preliminary microscopic evaluation of wet mount gill preparations confirmed the presence of bacteria suspected to be the causative agent of BGD. Mortality began to increase substantially 1 - 2 days prior to the initiation of treatment.

One day prior to the first chloramine-T treatment, 5 fish were captured and removed from each test unit for a pre-treatment fish health evaluation and confirmation of BGD. Gill tissue was collected, preserved, and processed according to standard procedures for histological examination (Sheehan and Hrapchak 1980), and stained gill squash slides were prepared. The gill squash procedure was repeated three other times during the course of the study and histological samples were collected again at the end of the study. Processed gill

## Results from a Chloramine-T Clinical Efficacy Trial

squashes were evaluated using a procedure to approximate level of uniformity of bacterial infection. Fifteen fields of view were observed at 400x for each of 5 fish from each test unit. Long filamentous bacteria were counted and numbers of gram-negative rod shaped bacteria were estimated. For graphing purposes, where bacteria were present in a field of view, regardless of number of bacteria, a value of 1 was assigned. Conversely, if no bacteria were present in the field of view a value of zero was assigned. A maximum total of 75 "points" was possible for each tank (5 fish x 15 fields of view).

Chloramine-T was administered to treated tanks on days 1, 3 and 5. Treatment dosage and duration was 12 mg/L for 60-minutes. A standing bath treatment method was used. Three aliquots of chloramine-T were weighed out and dissolved in 1 gallon of source (Penny Creek) water. Water flow to each tank was turned off and all tanks were given their respective treatment of chloramine-T or pure source water (for untreated controls), tank contents were stirred to ensure thorough mixing. After 60 minutes, water flow was turned back on to each tank and adjusted to 57 L/minute. Dissolved oxygen was monitored during the treatment period. If DO levels dropped below 6 mg/L, supplemental oxygen was added. During the course of treatment, a single rearing unit required supplemental oxygen.

A blinding technique was used to minimize data collection bias. Study participants involved in data collection, chloramine-T dose verification and fish health evaluations did not know which tanks received chloramine-T.

Mortality was the primary response variable. Dead fish were counted and removed from test units twice daily. Temperature and dissolved oxygen (DO) were measured twice daily. Water hardness and pH were measured at the beginning and end of the study. Chloramine-T treatment concentrations were verified using a DPD based chlorine test kit from Hach Company, Loveland, Colorado that measures free and total chlorine. Methodology used was developed by staff at the Upper Mississippi Science Center (V.K. Dawson, Biological Resources Division, USGS, personal communication). Water samples were taken midway through the 60 minute treatment period for chloramine-T dose verification. Samples were analyzed within one hour.

Fish were fed daily with Biodiet (Bioproducts, 1935 NW Warrenton Dr., Warrenton, Oregon) 1.0 mm at a rate of 3.0% body weight per day using a belt feeder set to deliver feed over an 8-h period. Amount of feed to be given to each tank was recalculated at least once per week. On treatment days, fish were not fed and tanks were cleaned prior to administering treatment. On non-treatment days, tanks were cleaned after removing dead fish in the morning.

Statistical tests were performed using SYSTAT (Wilkinson 1990). An independent t-test was used to detect differences between cumulative mortality among treated and untreated test units. Where differences are stated to be significant, a level of  $p \leq 0.05$  is implied.

### Results

The chloramine-T treatment used effectively controlled mortality among fall chum salmon caused by BGD. Mortality among treated test units at the end of the 19-d study was significantly lower than among the non-treated units. Total mortality among treated and untreated units ranged from 186 - 1,713 and 13,397 - 17,634, respectively (Table 2). Mean total mortality among treated and untreated units was 1,298 and 16,014, respectively.

Table 2. Total mortality and percent total mortality among all test units at the end of the 19-d study.

Tank No.	Treatment Condition	Total Mortality	% Cumulative Mortality
2	Treated	1,719	11.2
13	Treated	1,994	13.7
14	Treated	186	1.0
3	Control	17,012	94.7
4	Control	17,634	99.8
15	Control	13,397	98.3
Mean			
	Treated	1,298	8.6
	Control	16,014	97.6

At the end of the study, mean cumulative mortality among treated and untreated units was approximately 8.6% and 97.6%, respectively. Rate of mortality was extremely high among non-treated fish (Figure 1). By day 5 of the study, approximately 49.1% of the untreated fish had died while only 6.1% of the treated fish had died.

While cumulative mortality continued to mount among untreated test units, daily mortality among treated units peaked on day 5, the final day of chloramine-T treatment. Daily mortality among treated units decreased dramatically following the final treatment, and remained at basal levels for nearly the entire duration of the study. Cumulative mortality among treated units following the final treatment accounted for only an additional 2.5% of the total mortality, while an additional 48.5% mortality was observed among untreated test units.

A fish health evaluation of fish collected prior to the start of the study showed a heavy infection of long, filamentous bacteria of gills. Evaluation of the severity and uniformity of bacterial infection demonstrated a high level of infection among fish from 5 test units (bacteria was present in all 75 fields of view) and a moderate level of infection (bacteria was present in 16 fields of view) among fish in the remaining test unit. Figure 2 summarizes the presence of gram-negative, long filamentous bacteria throughout the study. One day after the final chloramine-T treatment, no long filamentous bacteria were observed on gills of fish from treated units. After the study had terminated, some long filamentous bacteria were again present on gills of fish from treated test units. The highest level of bacterial infection corresponded to the test unit which began to experience elevated mortality in the last few days of the post-treatment period. While presence of long, filamentous bacteria slowly decreased among fish from untreated test units, gills quickly became colonized with gram-negative, rod shaped bacteria. Gram-negative rod shaped bacteria were not only present in nearly every field of view of gills from untreated units, but by day 6 of the study each field of view revealed in excess of 500 bacteria. The increased combined gill bacteria correlates directly with increased mortality.

Evaluation of histological gill tissue samples collected before and after chloramine-T treatment show additional benefits to the therapy. Prior to treatment, gill epithelial tissue was swollen, and lots of debris, including bacteria, were observed between gill lamellae. Following treatment, gill tissue appeared normal, with only a thin

layer of epithelial tissue, and very little debris between lamellae. There appeared to be no damage to gill tissue as a result of chloramine-T exposure.

Due to mortality and fish growth, culture conditions such as flow index and density index changed over the course of the study. At the start of the study, there was no significant difference between numbers of fish in treated and untreated tanks. At the end of the study, flow and density index values were similar to what they had been before the study started among treated test units only. Among untreated units, these values were substantially lower. It was initially thought that as cumulative mortality mounted, culture conditions would improve and stressors related to the previously high flow and density index values would be diminished, culminating in recovery of untreated fish from the bacterial infection. This in turn would result in lessened mortality. It was further thought that because mortality was not excessive among treated tanks and fish continued to be reared under stressful conditions, that there would be a remission/reinfection and mortality would begin to climb during the post-treatment period. Neither of these concerns were realized within the 19-d study period.

The mean chloramine-T concentration among all treated tanks was 12.7 mg/L and ranged from 11.5 - 13.9 during the course of the study (Table 4). The overall mean measured value of chloramine-T among non-treated tanks was 0.18 mg/L and ranged from 0.0 - 0.8 mg/L. This was not suspected to be chloramine-T (or chlorine for that matter) but was probably an artifact or a result of turbidity in the water sample.

Mean water temperature and dissolved oxygen values over the course of the trial were approximately 10.0° C and 9.9 mg/L, respectively and were consistent throughout the study for all test units. Minimum and maximum daily water temperatures were 9.4° and 10.7° C, respectively. Minimum and maximum daily dissolved oxygen values were 7.3 - 11.5 mg/L, respectively. Mean DO saturation levels ranged from 70 - 98%. Mean water hardness and pH values, measured at the beginning and end of the study, were approximately 38 mg/L (as CaCO<sub>3</sub>) and 7.3 - 7.4, respectively. During the course of treatment, rearing unit #15 required supplemental oxygen. This unit also had the lowest DO levels throughout the entire study period. The mean DO level in this unit was approximately 7.3 mg/L, while mean DO levels in the other five units ranged from 8.6 - 9.6 mg/L. The unit

# Results from a Chloramine-T Clinical Efficacy Trial

Table 4. Mean chloramine-T concentrations from analyses of water samples taken during chemical treatment.

Tank #	2 Treated	3 Control	4 Control	13 Control	14 Treated	15 Treated	dup- licates
Target Concentration (mg/L)							
	12	0	0	0	12	12	12
Observed Concentration (mg/L)							
5/19/97	13.1	0.0	0.0	0.4	13.5	12.3	13.5
5/21/97	12.3	0.0	0.0	0.4	11.9	13.9	11.5
5/23/97	13.5	0.0	0.8	0.0	11.9	11.5	13.9
Mean Conc.	13.0	0.0	0.3	0.3	12.4	12.6	13.0
Std. Dev.	0.6	0.2	0.5	0.2	0.9	1.2	1.3
Coeff. of Variation	4.7%	n/a	n/a	n/a	7.3%	9.5%	10.0%
n/a - not applicable							

with the lowest DO levels had the highest number of fish at the start of the study, as well as the lowest total mortality at the end of the study. This higher biomass could account for the lower DO levels.

## Discussion

Chloramine-T was clearly effective in controlling mortality in fall chum salmon caused by BGD associated with flavobacters. Rate of mortality was extremely high among untreated test units. Had the study gone on for another week, chances are that no fish would have been left alive in the untreated units. On the other hand, mortality among fish treated with chloramine-T was controlled and had returned to basal levels during the post-treatment period. Mortality began to climb again in one of the treated units, but this was most likely as a result of stress caused by the high densities and low flow that were maintained during the course of the study. Mortality results for this particular test unit were supported by the bacteria presence/absence data. An increase in presence of long, filamentous bacteria were observed at about the same time mortality began to increase. This exemplifies the need to consider culture conditions when treating for environmentally induced diseases, such as BGD.

It has been recommended that therapeutic treatment be administered early in a disease outbreak, or prophylactically if it is expected to occur. A disease such as BGD, if either left untreated or failing to correct culture conditions can compromise the health of fish to the point where mortality may reach extreme levels quickly. Fish used in this study displayed behavior characteristic of BGD nearly a week before the start of the study. At the time the study started, fish had been diagnosed with a Grade I designation of hyperplastic severity (Post 1987) and mortality had become excessive. In spite of the delay in treatment from the first time BGD was suspected, chloramine-T therapy not only controlled mortality among diseased fish, but reduced swelling of gill lamellae and debris between gills and eliminated bacteria associated with the gills. Results such as these suggest that prompt treatment would have resulted in more profound control of mortality among treated fish.

A goal of most fish culturists/fish health biologists would include management of environmental and culture conditions conducive to minimizing the incidence of an epizootic altogether. However, when it is necessary to treat therapeutically, it is essential to have access to chemicals that are as effective as chloramine-T. Long-term access to chloramine-T will only come if this chemical is granted approval for use in aquaculture by FDA.



## Acknowledgments

I thank Ron Wong, Paul Kaiser, Bud Young and Dave Knox of the Quilcene National Fish Hatchery, and Joy Evered and Ray Brunson of the USFWS Olympia Fish Health Center for their assistance and expertise that made this study possible.

## References

- Brown, G.E. and R.J.F. Smith 1997. Conspecific skin extracts elicit antipredator responses in juvenile rainbow trout (*Oncorhynchus mykiss*). *Can. J. Zool.* 75:1916-1922.
- Brown, G.E. and R.J.F. Smith (In Press) Acquired predator recognition in juvenile rainbow trout (*Oncorhynchus mykiss*): conditioning hatchery reared fish to recognize chemical cues of a predator. *Can. J. Fish. Aquat. Sci.*
- Donnelly, W. A. and F. G. Whoriskey Jr. 1993. Transplantation of Atlantic salmon (*Salmo salar*) and crypsis breakdown. In R. J. Gibson and R. E. Cutting (editors), Production of juvenile Atlantic salmon, *Salmo salar*, in natural waters, p. 25-34. *Can. Spec. Publ. Fish. Aquat. Sci.* 118.
- Gotceitas, V. and J-G. J. Godin. 1993. Effects of aerial and in-stream threat of predation on foraging by juvenile Atlantic salmon (*salmo salar*). In R. J. Gibson and R. E. Cutting (editors), Production of juvenile Atlantic salmon, *Salmo salar*, in natural waters, p. 35-41. *Can. Spec. Publ. Fish. Aquat. Sci.* 118.
- Healy, M. C., and U. Reinhardt. 1995. Predator avoidance in naïve and experienced juvenile chinook and coho salmon. *Can. J. Fish. Aquat. Sci.* 52:614-622.
- Kanayama, Y. 1968. Studies of the conditioned reflex in lower vertebrates. X. Defensive conditioned reflex of chum salmon fry in a group. *Mar. Biol.* 2:77-87.
- Mathis, A. and R. J. F. Smith. 1993. Fathead minnows, *Pimephales promelas*, learn to recognize northern pike, Bills, T.D., L.L. Marking, V.K. Dawson, and J.J. Rach. 1988. Effects of environmental factors on the toxicity of chloramine-T to fish. U.S. Fish and Wildlife Service, Investigations in Fish Control 96, Upper Mississippi Science Center, P.O. Box 818, LaCrosse, Wisconsin.
- Bowker, J.D. and D.A. Erdahl. 1997. Observations on the efficacy of chloramine-T treatment to control mortality in a variety of salmonids. *Progressive Fish-Culturist*. In Press
- Bullock, G.L. 1990. Bacterial gill disease of freshwater fishes, Fish Disease Leaflet 84, U.S. Dept. of the Interior, Fish and Wildlife Service, Washington DC.
- Bullock, G. L., R. L. Herman, and C. Waggy. 1991. Hatchery efficacy trials with chloramine-T for control of bacterial gill disease. *Journal of Aquatic Animal Health* 3:48-50.
- Ferguson, H.W., V.E. Ostland, P. Byrne, and J.S. Lumsden. 1991. Experimental production of bacterial gill disease in trout by horizontal transmission and bath challenge. *Journal of Aquatic Animal Health* 3:118-123.
- From, J. 1980. Chloramine-T for control of bacterial gill disease. *The Progressive Fish-Culturist* 42:85-86.
- Lasee, B.A., Ed. 1995. Introduction to fish health management, 2<sup>nd</sup> edition. U.S. Department of the Interior, Fish and Wildlife Service. LaCrosse Fish Health Center, Onalaska, Wisconsin. 139 pp.
- Lumsden, J. S., V. E. Ostland, D. D. MacPhee, J. Derksen, and H. W. Ferguson. 1994. Protection of rainbow trout from experimentally induced bacterial gill disease caused by *Flavobacterium branchiophilum*. *Journal of Aquatic Animal Health* 6:292-302.
- Post, G. 1987. Textbook of fish health, revised. TFH Publications, Inc., Neptune City, New Jersey. 288 pp.
- Sheehan, D.C. and B.B. Hrapchak. 1980. Theory and Practice of Histotechnology, 2<sup>nd</sup> edition. The C.V. Mosby Co., St. Louis, Missouri. 481 pp.
- Wakabayashi, H, G.J. Huh and N. Kimura. 1989. *Flavobacterium branchiophila* sp. nov., a causative agent of bacterial gill disease of freshwater fishes. *International Journal of Systematic Bacteriology* 39:213-216
- Wilkinson, L. 1990. SYSTAT: The system for statistics. SYSTAT Inc. Evanston, IL 677 pp.

## **The Use of Penicillin-G for Control of Bacterial Coldwater Disease in Salmonid Fishes**

William T Cox <sup>1</sup> and Tresa Veek  
*California Department of Fish and Game*

Abstract— Bacterial cold-water disease (BCWD) caused by *Flavobacterium psychrophilum* is a significant pathogen in the Pacific Northwest. High rates of mortality in steelhead and rainbow trout fry have occurred in California hatcheries for the last three years. Adequate therapies for control exist but are not currently legal due to FDA regulations on the use of medicinal compounds on food fishes. Recent legislation (AMDUCA 1994) allowing relaxation of regulations concerning veterinary extra-label use of registered antibiotics has allowed the use of "new" compounds for treatment of this difficult disease. During the winter and spring of 1997, California has conducted several tests of Penicillin-G, both prophylactically and therapeutically, to control BMD. Results and use patterns will be discussed at this meeting.

---

<sup>1</sup> Bill was only able to submit an abstract for publishing in the Proceedings because his computer was stolen and he was not able to recreate his data.



Cox and Veek

# Culling of Eggs from BKD Positive Spring Chinook in Oregon.

Tony Amandi and Leslie Smith

Oregon Department of Fish and Wildlife, Department of Microbiology, Nash 220,  
Oregon State University, Corvallis, Oregon 97331-3804

Spring chinook salmon are highly susceptible to Bacterial Kidney Disease (BKD) caused by the Gram positive rod *Renibacterium salmoninarum*. Over the years, outbreaks of this disease have occurred at all Oregon Department of Fish and Wildlife hatcheries which raise spring chinook salmon (*Oncorhynchus tshawytscha*). We have come to rely on the use of oral erythromycin medicated food and prespawning injections of adult fish to treat for the disease. Treatments have been administered either prophylactically as attempts to prevent clinical disease, or therapeutically where outbreaks of BKD have occurred.

In some cases we have seen outbreaks of BKD occurring in the fall and winter in spite of two prophylactic erythromycin feedings given to spring chinook. A culling program to remove eggs from BKD positive parents began in 1993 with Willamette River stock spring chinook salmon and was expanded over the past four years to encompass all the Willamette River system stocks and a Deschutes River stock. Except for the Willamette stock 1993 brood where both males and females were sampled, only females have been tested for *R. salmoninarum*. The culling was initiated as an attempt to decrease the BKD outbreaks during rearing and reduce the level of BKD in smolts. In conjunction with the elimination of eggs from BKD positive fish, a reduction in the amount of erythromycin being fed prophylactically at some of our facilities has resulted in substantial cost savings.

Fish were designated positive for BKD either by visual signs of disease (pustules or swollen kidneys) or by examination of kidney tissue using the enzyme linked immunosorbent assay (ELISA) technique. Numbers of fish which were positive for *R. salmoninarum* ranged from a low of 1.4% in the Willamette River stock in 1994 to a high of 47.8% in the North Santiam River stock in 1997. Eggs from positive fish were culled at all facilities to varying degrees. Where possible, 100% of eggs from females which had kidney pustules or from which *R. salmoninarum* antigen was detected by ELISA were discarded. At other times, due to production constraints, the number of culled eggs was as low as 30%. In cases where fish from *R. salmoninarum* positive females were reared we have segregated those fish and in some cases increased the number of prophylactic erythromycin feedings.

Besides the dollar savings due to reduced erythromycin feedings, we have noted either an absence or a delay of BKD outbreaks in populations where progeny originated from females where no *R. salmoninarum* antigen was detected. An outbreak of BKD in culled negative fish has occurred only in the McKenzie River stock. These fish are held in a facility where *R. salmoninarum* is highly prevalent in the water supply. In this case the outbreak was delayed by two months and the incidence and mortality was less severe than in the past. We will continue culling in these stocks as long as we see reduced levels of BKD in the rearing juveniles. In time, we will be able to find out whether this also affects the *R. salmoninarum* positive number of returning adults.

## Introduction

Bacterial kidney disease (BKD) caused by the Gram positive bacterium *Renibacterium salmoninarum* affects many species of salmonids. Spring chinook salmon (*Oncorhynchus tshawytscha*) are particularly susceptible to the organism. The Oregon Department of Fish and Wildlife (ODFW) has attempted to control this disease with the use of erythromycin. Returning adults are injected with erythromycin at a target of 22 mg/Kg of body weight from once to three times during the summer holding period. Juvenile fish have been prophylactically or therapeutically fed erythromycin medicated feed (Gallymicin and more recently Aquamycin) at a target dose of 100 mg/Kg of body weight from one to three times during the rearing period.

In spite of attempts to reduce the impact of *R. salmoninarum* on spring chinook salmon stocks in Oregon's Willamette River system, outbreaks of the disease continued to occur with regularity at several ODFW facilities. To further reduce the effect of *R. salmoninarum* on these stocks, a culling program began in 1993 with Willamette River stock spring chinook and has been expanded over the last four years to include all the Willamette River system stocks and a Deschutes River stock. The culling program was initiated as an attempt to reduce the BKD outbreaks during rearing and decrease the level of *R. salmoninarum* in migrating smolts. As part of the culling program it was also proposed to reduce the reliance on erythromycin for certain stocks, thus reducing medication costs.

## Materials and Methods

The culling program has occurred at the following Willamette River system facilities: Willamette Hatchery (Willamette River stock, 1993-1997); McKenzie Hatchery (Willamette River stock, 1994-1997 and McKenzie River stock, 1995-1997); Minto Pond (North Santiam River stock, 1996-1997); South Santiam Hatchery (South Santiam River stock, 1996-1997) and Clackamas Hatchery (Clackamas River stock, 1997). In addition, the Deschutes River stock was sampled in 1997 at Round Butte Hatchery.

The enzyme linked immunosorbent assay (ELISA) method was used for detection of *R. salmoninarum* antigen. Except for the 1993 brood Willamette River stock where both males and females were sampled, only females were sampled for the presence of *R. salmoninarum* antigen. Kidney samples (approximately 2 g) were collected from fish with individual razor blades and placed in whirl pack bags. Eggs were placed in incubator trays labeled with the same number as the whirl pack bags. The samples were weighed and diluted 1:4 with phosphate buffered saline and homogenized by running a rolling pin over the bags several times. Aliquots were poured into tubes, boiled and centrifuged. The supernatant was then used to load 96 well plates precoated with goat anti *R. salmoninarum* antigen. The plates were incubated, washed, reacted with a secondary anti-body conjugated to horse radish peroxidase and followed by ABTS (a chromagen) and peroxidase as a color substrate. Positive samples change color. Then plates are read and the results downloaded to a computer. We assigned the following ELISA optical density (OD) readings to *R. salmoninarum* antigen levels:  $<0.1$  = negative;  $0.1 - 0.199$  = low;  $0.2 - 0.499$  = moderate and  $\geq 0.5$  = high.

Where possible, eggs from fish with OD readings of  $\geq 0.1$  were destroyed. In situations where not enough negative eggs were available for production targets, eggs from low level positive adults were segregated and fish were reared separately from those which came from negative adults. In some of these cases the fish were fed erythromycin as Aquamycin medicated feed three times during the rearing period. In other cases, the use of erythromycin was reduced by giving fish a single feeding in the spring and eliminating the summer treatment. Fish at McKenzie Hatchery continue to receive two erythromycin feedings because of horizontal transmission from wild and feral fish in its water supply.

## Results and Discussion

Spring chinook were found to have *R. salmoninarum* antigen in kidney tissue at a 10.6% level during the first five years of the program. Percent of fish with detectable antigen ranged from low levels of 1.4% (Willamette River stock, 1994) and 2.0% (Clackamas River stock, 1997) to high levels of 47.8% (North Santiam River stock, 1997) and 44.6% (Deschutes River stock, 1997) (Table 1).

Of all the fish which tested positive for *R. salmoninarum* antigen between 30 and 100% of their eggs were destroyed. The remaining eggs and resulting fry were segregated from those testing negative. All eggs from females which tested positive at moderate and high levels were destroyed and only eggs from those testing at low levels were kept as needed to reach production targets (Table 2).

Of these, the 1995 McKenzie River stock was fed Aquamycin three times before being released in the fall of 1996 for migration to the ocean. No clinical BKD was detected in these fish. Other groups were segregated and fed Aquamycin two times with no clinical BKD detected during rearing until release in the spring of 1997.

Overall, 58% of the fish which tested positive for *R. salmoninarum* antigen had low level OD readings while 11% had moderate and 31% had high readings (Figures 1 and 2, Table 3). In most cases, the majority of the fish had low levels, followed by high and moderate levels of antigen. One third of the fish in the high OD level group had clinical signs of BKD at spawning time. These fish had typical kidney pustules or gray or swollen kidneys. The eggs were discarded, no kidney samples were collected and the fish were assumed to have high OD readings.

The percent of eggs culled by stock and year are presented in Table 4. Eggs which were removed from the population came from all parts of the spawning cycle. In 1997 in the Willamette and McKenzie stocks there was no correlation between the presence of *R. salmoninarum* antigen and erythromycin injections. Positive fish were as likely to come from non injected as well as injected females. During the program, 10.9% or 4,984,600 eggs have been culled due to the presence of *R. salmoninarum* antigen in kidney tissue of the adults or because eggs

## Culling of Eggs from BKD Positive Spring Chinook in Oregon

from fish whose kidneys tested negative were mixed together in a tray with those of a female testing positive. This accounted for 1,834,650 eggs being destroyed which would have been considered negative for the presence of the antigen (Table 4). Starting in 1997 eggs from each female were separated in each tray by a plastic divider at all but one facility. This step eliminated the need to remove eggs from fish considered to be negative for *R. salmoninarum* antigen.

Outbreaks of BKD have not occurred in the Willamette, North Santiam and South Santiam river stocks since the culling program began. Low if any signs of clinical BKD have been detected in these stocks during this period. An outbreak of BKD did occur on McKenzie River stock at McKenzie Hatchery in the negative 1995 brood fish. This outbreak was delayed by two months from annual outbreaks and was of much lower impact in numbers of fish affected.

Erythromycin feedings have been decreased to once for the Willamette, North Santiam and South Santiam River and Clackamas River stocks. The McKenzie River and Deschutes River stocks will continue to receive two feedings of medicated feed. In 1998, the Clackamas River stock and the negative North Santiam River stock will

receive no medicated feed as we further wean ourselves from reliance on erythromycin to control BKD at locations where the pathogen is not a problem in our water supplies.

### Acknowledgments

The following agencies provided funds for this program: U. S. Army Corps of Engineers, Bonneville Power Administration and Portland General Electric.

Thanks to the crews of the following hatcheries for their patience and invaluable help: Clackamas, Marion Forks, McKenzie, Round Butte, South Santiam and Willamette.

The following people have participated in this program over the years: Craig Banner, Mary Edwards, Judy Engelking, Mark Engelking, April Erickson, Rich Holt, Nadine Hurtado, John Kaufman, Terry Kreps, Harriet Lorz, Tim Plawman, Mark Redhead, Greg Rutherford, Bryant Spellman, Sharon Vendshus and Robin Whitmore.

Special thanks to Harry Lorz for his support over the five years of the culling program for the Willamette River spring chinook stocks.

Table 1. Number of fish positive for *Renibacterium salmoninarum* in six Oregon stocks of spring chinook salmon from 1993-1997.

Fish Stock	Brood Year	Total Fish Sampled	BKD Positive Fish	Percent BKD Positive
All	1993-1997	10,190	1,082	10.6
Willamette	1993	1,454	172	11.8
Willamette	1994	852	12	1.4
Willamette	1995	1,097	80	7.3
Willamette	1996	1,351	93	6.9
Willamette	1997	1452	170	11.7
McKenzie	1995	472	44	9.3
McKenzie	1996	544	27	5.0
McKenzie	1997	622	64	10.3
North Santiam	1996	193	58	30.0
North Santiam	1997	224	107	47.8
South Santiam	1996	642	74	11.5
South Santiam	1997	524	28	5.3
Clackamas	1997	440	9	2.0
Deschutes	1997	323	144	44.6

Table 2. Culled eggs due to presence of *R. salmoninarum* in adult spring chinook salmon stocks from 1993 to 1997.

Fish Stock	Year	Total BKD fish	Low BKD # - % culled	Mod. BKD # - % culled	High BKD # - % culled	Total % culled
Willamette R.	1993	172	88 — 35	24 — 100	60 — 100	80
Willamette R.	1994	12	5 — 100	0	7 — 100	100
Willamette R.	1995	80	42 — 100	7 — 100	31 — 100	100
Willamette R.	1996	93	58 — 100	7 — 100	28 — 100	100
Willamette R.	1997	170	130 — 100	7 — 100	33 — 100	100
McKenzie R.	1995	44	21 — 0	2 — 100	21 — 100	30
McKenzie R.	1996	27	13 — 100	5 — 100	9 — 100	100
McKenzie R.	1997	64	48 — 100	5 — 100	11 — 100	100
No. Santiam R.	1996	58	18 — 22	2 — 100	38 — 100	93
No. Santiam R.	1997	107	75 — 65	19 — 100	13 — 100	46
So. Santiam R.	1996	74	23 — 100	14 — 100	37 — 100	100
So. Santiam R.	1997	28	23 — 100	2 — 100	3 — 100	100
Clackamas R.	1997	9	4 — 100	0	5 — 100	100
Deschutes R.	1997	144	83 — 83	25 — 100	36 — 100	48

# Culling of Eggs from BKD Positive Spring Chinook in Oregon

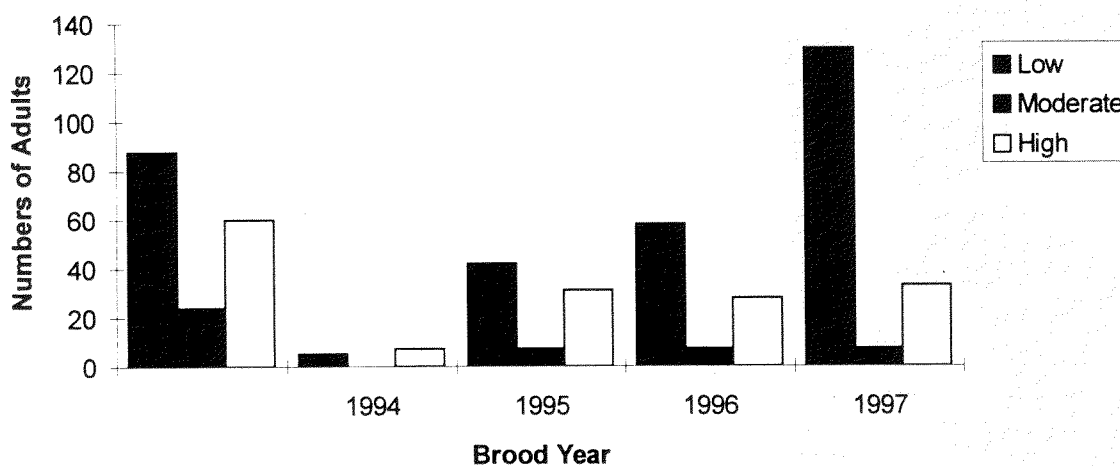
Table 3. Levels of *R. salmoninarum* in six Oregon adult spring chinook stocks from 1993 to 1997.

Fish Stock	Year	Total fish <sup>1/</sup>	Low BKD # - % positive	Mod. BKD # - %positive	High BKD # - %positive	Clinical BKD <sup>2/</sup>
Willamette	1993	1454	88 — 6.1	24 — 1.7	60 — 4.1	20
Willamette	1994	852	5 — 0.6	0	7 — 0.8	2
Willamette	1995	1097	42 — 3.8	7 — 0.6	31 — 2.8	7
Willamette	1996	1351	58 — 4.3	7 — 0.5	28 — 2.1	8
Willamette	1997	1452	130 — 9.0	7 — 0.5	33 — 2.3	19
McKenzie	1995	472	21 — 4.5	2 — 0.4	21 — 4.5	11
McKenzie	1996	544	13 — 2.4	5 — 0.9	9 — 1.7	1
McKenzie	1997	622	48 — 7.7	5 — 0.8	11 — 1.8	5
N. Santiam	1996	193	18 — 9.3	2 — 1.0	38 — 19.7	22
N. Santiam	1997	224	75 — 33.5	19 — 8.5	13 — 5.8	2
S. Santiam	1996	642	23 — 3.6	14 — 2.2	37 — 5.8	8
S. Santiam	1997	524	23 — 4.4	2 — 0.4	3 — 0.6	1
Clackamas	1997	440	4 — 0.9	0	5 — 1.1	4
Deschutes	1997	323	83 — 25.7	25 — 7.7	36 — 11.2	0

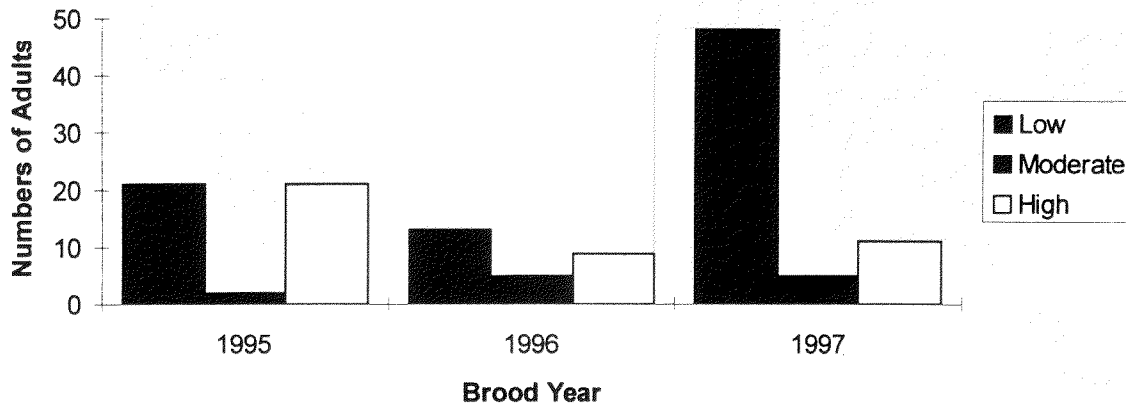
<sup>1/</sup> Includes fish culled at spawning time due to presence of clinical BKD signs.

<sup>2/</sup> Fish with typical BKD kidney pustules, no samples collected. These fish were counted as high level positives.

Figure 1. Levels of BKD on Willamette Stock Spring Chinook



**Figure 2. Levels of BKD on McKenzie River Stock Spring Chinook**



**Table 4. Percentage of BKD positive eggs culled from six Oregon stocks of spring chinook salmon from 1993 to 1997.**

Fish Stock	Brood Year	Number of Eggs Culled	Percent of Eggs Culled
All	1993 - 1997	4,984,600	10.9
Willamette	1993	625,000*	15.0
Willamette	1994	54,000*	0.7
Willamette	1995	616,500*	11.5
Willamette	1996	738,000*	12.5
Willamette	1997	1,152,000*	19.0
McKenzie	1995	96,800*	5.1
McKenzie	1996	214,200*	10.4
McKenzie	1997	267,200	9.5
North Santiam	1996	200,600*	24.9
North Santiam	1997	251,000	22.6
South Santiam	1996	379,900*	13.0
South Santiam	1997	126,000	4.8
Clackamas	1997	40,500	2.0
Deschutes	1997	222,000	21.3

\* Approximately 45% of these eggs were from negative females but were destroyed because they were sharing a tray with those of a positive female.

# Integrated Management of Bacterial Kidney Disease at IDFG Lower Snake River Compensation Fish Hatcheries

A. Douglas Munson

*Eagle Fish Health Laboratory, Idaho Dept. Of Fish and Game*

Abstract— Since 1993, IDFG has applied an aggressive disease management program to control Bacterial Kidney Disease (BKD) in the chinook hatchery program. This program includes intraperitoneal injections of erythromycin, iodophor disinfection of eggs, 100% ELISA examination of brood females, ELISA-based segregation/culling of high BKD progeny, prophylactic erythromycin-medicated feed treatments, and segregated releases of high BKD groups. This program has resulted in limiting clinical BKD to high BKD segregation groups (with one exception). IDFG plans to use these ideas as a dynamic program and will explore different ELISA break-off points and different treatment strategies to effectively and economically control *Renibacterium salmoninarum*.

## IDFG Program to Limit BKD

Strategy	
1.	Intraperitoneal injections of erythromycin (20 mg/Kg body weight).
2.	Iodophor disinfection of eggs.
3.	100% ELISA testing of female brood stock.
4.	ELISA based segregation to limit horizontal transmission.
5.	Prophylactic feeding of erythromycin medicated feed.
6.	Segregated release of smolts.

## Conclusion

Idaho Department of Fish and Game hatcheries have the capability to control Bacterial Kidney Disease. This capability also limits horizontal transmission of *Renibacterium salmoninarum* to healthy hatchery fish

and probably wild fish as well, thus enhancing the chances of smolt survival and adult returns. There has no been an epizootic of BKD in low BKD segregation groups in any IDFG hatchery since 1993. In 1993, IDFG implemented 100% ELISA sampling of chinook brood



Munson

Historical Data for McCall Hatchery <sup>1</sup>

Brood Year	Brood	Juvenile	Pre-liberation
1987 <sup>2</sup>	44/187	37/288	28/60
1988 <sup>3</sup>	27/236	4/181	0/60
1989	4/60	0/131	0/60
1990	0/60	0/100	13/60
1991	19/60	0/50	0/60
1992	7/60	15/44	3/60
1993 <sup>4</sup>	136/515	1/89	0/20
1994	22/141	0/44	0/20
1995	37/58	0/40	0/40
1996	1/12	0/30	-----

Historical Data for Rapid River Hatchery <sup>5</sup>

Brood Year	Brood	Juvenile	Pre-liberation
1987 <sup>6</sup>	-----	27/174	24/120
1988 <sup>7</sup>	24/60	24/106	0/60
1989	18/60	0/112	0/60
1990	31/60	0/101	3/60
1991	10/60	0/32	18/180
1992	132/232	0/72	5/60
1993 <sup>8</sup>	685/1590	1/95	0/20
1994	57/120	0/68	0/20
1995	32/36	0/65	0/20
1996	288/327	0/20	-----

<sup>1</sup> Prior to BY93 results via DFAT. BY93 - BY96 brood testing via ELISA; juvenile via DFAT.

<sup>2</sup> BY87: Adult injections only.

<sup>3</sup> BY88 - BY92: Adult injections and 2x prophylactic erythromycin medicated feedings.

<sup>4</sup> BY93 - BY96: Adult injections, 2x erythromycin medicated feedings and segregated rearing and culling.

<sup>5</sup> Prior to BY93 results via DFAT. BY93 - BY96 brood testing via ELISA; juvenile via DFAT.

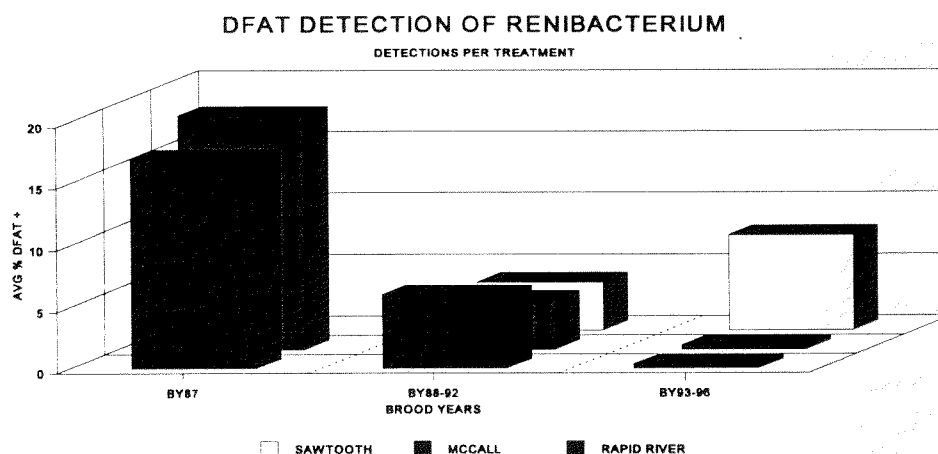
<sup>6</sup> BY87: Adult injections only.

<sup>7</sup> BY88 - BY92: Adult injections and 2x prophylactic erythromycin medicated feedings.

<sup>8</sup> BY93 - BY96: Adult injections, 2x erythromycin medicated feedings and segregated rearing and culling.

### Historical Data for Sawtooth Hatchery <sup>9</sup>

Brood Year	Brood	Juvenile	Pre-liberation
1987 <sup>1010</sup>	-----	-----	-----
1988	-----	-----	-----
1989 <sup>1111</sup>	2/5	4/235	0/80
1990	119/497	28/154	3/120
1991	63/203	2/117	0/80
1992	95/272	1/142	0/55
1993 <sup>1212</sup>	35/89	20/90 <sup>13</sup>	3/40 <sup>14</sup>
1994	9/12	0/35	0/40
1995	3/5	0/13	0/80
1996	9/15	-----	-----



BY87: Erythromycin Injection Only  
 BY88 - BY92: Erythromycin Injection + 2 Fry Feedings  
 BY93 - BY96 Erythromycin Injection + 2 Fry Feedings + Segregation

<sup>9</sup> Prior to BY93 results via DFAT. BY93 - BY96 brood testing via ELISA; juvenile via DFAT.

<sup>10</sup> BY87 - BY88: No data.

<sup>11</sup> BY89 - BY92: Adult injections and 2x prophylactic erythromycin medicated feedings.

<sup>12</sup> BY93 - BY96: Adult injections, 2x erythromycin medicated feedings and segregated rearing and culling.

<sup>13</sup> This DFAT positive represents part of the last epizootic of BKD in IDFG hatchery production fish. IDFG has had only two epizootics in high BKD segregation groups.

<sup>14</sup> This DFAT positive represents part of the last epizootic of BKD in IDFG hatchery production fish. IDFG has had only two epizootics in high BKD segregation groups.

Munson



# Session V

## **Captive Broodstock**

Session Chair:

Mike Delarm  
(National Marine Fisheries Service)

## Cryopreservation of Salmonid Sperm

Loren Jensen <sup>1</sup>

*Oregon Department of Fish and Wildlife*

What is cryopreservation? Basically, it is the long term storage of material at extremely low temperature.

Why do we use cryopreservation? We use cryopreservation for genetic conservation of existing fish stocks. With the thought of losing genetic diversity in specific native stocks the establishment of a program for the long term storage of fish germ plasma would serve as a insurance program.

I will show:

- 1) The preparation of the process involved in actual cryo-preservation procedures;
- 2) The thawing and reactivation of sperm for fertilization of eggs.

---

<sup>1</sup> Loren elected to only have the abstract of his talk published in the Proceedings.

Jensen

# Synchronized Spawning of Wild and Captive Broodstock

Jim Powell, Ph.D.

*Aquamatrix Research Ltd., 204-2527 Beacon Ave.  
Sidney, B.C., Canada, V8L 1Y1*

**Abstract-** In the natural course of salmonid maturation, environmental cues are translated into physiological change. This change is progressive and involves a cascade effect of hormones initiated in the hypothalamus to the pituitary to the gonad. Salmon have periods of susceptibility whereby the handling of maturing fish may cause a cessation of the maturation process. As well, maturation will occur at different rates in a large population and therefore fish will spawn at different times. This serves to spread out the spawning season over a period that is both species and strain specific. In some cases, spawning dates may be asynchronous in races of fish that have limited returns to natal streams. The results are similar: fisheries managers are left with stocks that are not mature and will not synchronously mature. Under INAD and ESC (Canada) approval we have developed a method to advance and synchronize maturation in wild and captive broodstock. In a case study, coho salmon and seawater rainbow trout were induced to mature using an implant placed in the dorsal sinus of the fish. In treated fish, spawning dates were significantly ( $P < 0.05$ ) advanced, milt production and viability was significantly increased and fry were significantly quicker to first feeding. There were no differences in mortality, egg size, fertilization rate or physical characteristics within the stocks. In three INAD and three ESC trials, this method has proven both safe (for humans and fish) and is effective.

## Overview of Spawning Endocrinology

In the maturation of salmonids, there is a complex interchange between the external and internal environments that initiates the maturation process. In terms of external cues, rates of change in photoperiod and absolute day length are the main influences with other factors such as temperature, nutrition and social effects all following in importance. The cumulative effect of external cues and positive response from self to mature is an initiation of physiological maturation led by the endocrine system.

The perception of environment through sight, smell, taste, salinity and touch is coordinated through higher brain centers. Through neural relay, external cues are coordinated with internal cues into a response to mature or not. If the signal to mature is positive, inhibition of signals and or a stimulatory response to proceed with maturation is received in a small area at the underside of the brain called the hypothalamus. The hypothalamus is the coordination center for higher neural inputs with direct links to the major hormone producing organ the pituitary. In response to stimulation from the hypothalamus, the pituitary manufactures and releases hormones that in turn initiate a further cascade of hormone responses from the gonads and other organs. The end result is a multiplication of hormonal and physiological responses that culminate in spawning.

### *Gonadotropin-Releasing Hormone (GnRH)*

In the case of reproduction, the hypothalamus releases gonadotropin-releasing hormone (GnRH) directly onto gonadotrophs located in the pituitary. GnRH was originally called LHRH because of the luteinizing

hormone releasing ability in rats. It was shortly thereafter disclosed that LHRH also released follicle-stimulating hormone (FSH). Further, there are nine known forms of GnRH in other vertebrates. However, the misnomer of LHRH persists. Despite nomenclature, GnRH has the effect of gonadotropin (GtH) liberation and synthesis on gonadotrophs.

### *Gonadotropic Hormones (GtH)*

In early maturation, the first GtH to be released is called GtH I. Later in the maturation process, another GtH called GtH II is similarly released in response to GnRH release. As GtH I levels fall towards the latter stages of maturation, GtH II levels increase in titre. Both GtH I & II are liberated to the systemic circulation and has the major effect of causing maturation of the gonad.

### *Steroids*

In salmon, GtH I effects the gonad to produce steroid hormones. Under the influence of GtHs the ovary and testis produce testosterone. In the ovary, this is converted to estradiol that is released into the bloodstream and is removed by the liver. In the liver, estradiol serves to induce the formation of yolk platelets that are likewise released into the bloodstream. The ovary removes the yolk platelets from the blood and incorporates the platelets into the egg. This is vitellogenesis and yolk deposition. In the testis, testosterone directly influences the development of milt.

Through this chain of endocrine events environmental cues are translated into a physiological process that results in the production of viable gametes. As fish culturists, we depend on this process. We also depend on

this process to perpetuate fish survival. However, this process is not infallible and captive fish will sometimes not spawn.

### **Synchronized Spawning**

Salmonids and other captive fish can be induced to spawn either as individuals or as a population. This practice can alleviate the problems of the following scenarios:

#### *Scenario 1:*

In a large population of returning or captive salmonids, the maturation date of the population is a predictable event. Normally, stocks have a history to their spawning season that is marked by the date of first spawning, median and last spawning. The distribution of spawners over time usually takes the shape of a normal distribution or a bell curve. At the onset of spawning, some fish will mature before others in small numbers. The bulk of the population spawns within a finite period followed by a few fish that spawn towards the end of the season. There are differences in spawning times for species. In addition, there are differences in spawning times within a given stock and even race. The environment, behavior of the animals and physiological factors can contribute to altering spawning dates from year to year and stock to stock. The end result is a loosely-defined spawning period for a stock that if large in number takes the form of a bell curve that may start and end within a predictable time, but with a period of grace.

In a threatened or endangered population the distribution of spawning times for the former, larger population will be the same; some fish spawn early, some late. However, the total numbers of returns may be greatly reduced. In some cases, only a few individuals represent the former population. These fish may not be synchronized in their spawning times; some will spawn early, some late. In these populations, it is important for the preservation of genetic material that all fish spawn synchronously.

To preserve the stock, either males or females must be made to spawn at controlled times. To help the situation, it would be an asset if males can be made to produce milt for longer periods or if females and males could be made to spawn earlier. This synchronization of spawning dates can be achieved through induced maturation.

#### *Scenario 2:*

In some locations, broodstock are seined or captured in seawater at the river mouth or below the hatchery. In these circumstances using a sedative (not an anesthetic) such as Marinil, working quickly and gently minimizes stress of transport. Despite these best efforts male salmon which were running with milt before transport have dried up in the hatchery. As well, female salmon that were soft before capture sometimes harden up after transport. This transport shock may, in part, be due to high cortisol levels induced by transport stress. These elevated cortisol levels may have a negative feed-back on GnRH production which in turn stalls the maturation process. The result is fish that stop maturing when transported to the hatchery.

In addition to the above effects, handling shock may cause high mortality in freshwater, reduced fecundity and a higher incidence of abnormal eggs. To the hatchery staff, it would be preferred that the fish were put back on track and restart maturation in order to minimize the effects of transport. Induced maturation can help restart a stalled maturation process.

#### *Scenario 3:*

Periodically, stocks with high escapement will return in large numbers. In this case there is a need to capitalize on the abundance of genetic diversity that has been provided. As one to one matings are preferred to pooled or multiple crosses, it demands large numbers of ripe males and females to be made available at one time. In addition to breeding plans, it is desirable to use all the available stock that comes into the hatchery in a short time for two reasons: 1) the sooner a raceway or channel is cleared of fish, the faster it can be filled again, and 2) by removing spawners from the hatchery at a fast rate, the incidence of disease transmission from latent fish to new arrivals decreases. Further, most managers like to fill their trays as fast as possible to keep downstream schedules such as picking, ponding and first feeding moving at an optimal pace. In this case, synchronized spawning through induced maturation helps to provide a maximum amount of gametes at a controlled rate in a shorter period of time. The effect is a compressed spawning season that provides the optimal reproductive potential from a stock.

In each of these scenarios synchronized spawning by induced maturation contributes to the different objects of the specific problems: disparate maturation time, post-



## Synchronized Spawning of Wild and Captive Broodstock

transport shock and compressed spawning season. The methods of induced maturation vary from environmental controls such as photoperiod, to fish extracts such as acetone dried pituitaries, to purified or synthesized hormones. The essence of each of these methods is the same; the hormonal control of maturation is manipulated.

### Induced Maturation

Methods of controlling maturation by environmental conditions such as photoperiod require longer periods of time in captivity. For many, this is not possible. Rather, most fish are induced to mature by application of hormones. In these cases, exogenous hormones serve to copy or mimic the natural progression of hormone release, but at a determined time or rate. All methods seek to control reproduction at the level of the:

- ▶ Gonad through steroids,
- ▶ Pituitary through GtHs, and
- ▶ Hypothalamus through GnRH.

Common experimental methods use gonadal steroids for induced maturation, but this is not feasible for work in the hatchery. As well, this level of control is at the end of the endocrine chain that may leave little flexibility for secondary hormonal effects such as metabolic changes. Pituitary extracts or freeze-dried tissues have little or no quality control when it comes to protein hormone content. That is, extracts and tissue preparations have undetermined hormone content and dose is not uniform from use to use. As well, with crude extractions there are a multiplicity of other pituitary hormones that may be present in the extract that have no bearing on reproduction, but may influence other physiological functions. These factors considered results are predictable, but may be variable or not repeatable when using extracts or tissues.

In contrast to steroids or impure products for induced maturation, purified or synthetic hypothalamic hormones pose a positive alternative. GnRH peptides are small and can be synthesized in a pure form. Analogues of the peptides are up to 100 times more potent than native forms. As well, some forms found in fish are not found in humans and have reduced potency in humans. However, in fish, analogues are potent and cause the desired effect of induced maturation within days to weeks.

GnRH peptides uses the fish's own chain of hormonal events to:

- ▶ Advance and extend the maturation process,
- ▶ Restart stalled maturation, and
- ▶ Compress and synchronize the spawning season.

GnRH can be administered once in a biodegradable, natural vehicle through a sterile procedure. The process is user friendly and safe for fish and humans. When administered as a pellet implant, GnRH preparations can be used to advance the normal spawning time by 6-8 weeks. When used inside the normal spawning season, commercial preparations of GnRH can cause spawning within 10 days post use. These treatments have been shown to have no effect on fecundity or viability of eggs and increase the milt volume and length of time milt is made.

GnRH implants have been used for over 15 years to induce maturation in salmon. Products for commercial application are now available under both INAD (USA) and ESC (Canada) application. User-friendly, safe application methods have been used in trials both in the USA, Canada, the UK and in South America. The following case studies bear out the usefulness and applicability of GnRH implants.

### Case Studies

#### Coho-

##### Objective:

To advance ovulation date and compress the spawning season through induced ovulation using GnRH hormone implants and liquid GnRH hormone preparation.

##### Design:

25 males, 25 females, all held in FW

##### 5 Groups:

- 1) implanted with pellets,
- 2) implants and liquid hormone booster when gravid,
- 3) liquid hormone at first sign of spawning,
- 4) sham control, and
- 5) no treatment.

Fish were treated two to three weeks prior to the onset of the spawning season. Evaluation parameters were ovulation date, egg volume (number), egg survival (to

eyed and hatch) and time to first feeding. Tukey's and Dunn's tests were used on ranked data for non-parametric values. Significance was accepted at 95%.

**Results:**

There were no differences between controls and experimental groups for dam size, egg number, survival to hatching and survival to first-feeding. Experimental fish spawned in advance of controls by 3-4 weeks and within a week of first to last spawner. Time to first feeding was faster in offspring from treated fish. In males, milt volume was greater and groups 1 & 3 had higher sperm counts per ml than sham implanted controls.

**Seawater trout-**

**Objective:**

To advance ovulation date and compress the spawning season through induced ovulation using GnRH hormone implants and liquid GnRH hormone preparation.

**Design:**

25 males, 25 females matured in FW.

Two groups:

- 1) GnRH implants and
- 2) sham controls.

Fish were treated four weeks prior to the onset of the spawning season. Evaluation parameters were ovulation date, egg volume (number), egg survival (to eyed and hatch) and time to first feeding. Tukey's and Dunn's tests were used on ranked data for non-parametric values. Significance was accepted at 95%.

**Results:**

The results were the same for treated vs. non-treated fish in the coho experiment although the time of implanting was one week earlier than for coho salmon.

**Yukon chinook-**

Yukon River chinook salmon are sensitive to handling prior to spawning. When caught in the river and transported to the hatchery, Yukon River chinook will stall in the maturation process.

**Objective:**

To restart or ensure maturation in transported Yukon River chinook by using GnRH implants.

**Design:**

25 out of approximately 100 females were implanted with GnRH pellets 2 weeks prior to the normal spawning season.

**Results:**

12 of 25 fish spawned 21 days post implant. Five of the non-treated fish spawned in the same period.

**Atlantic Salmon-**

Atlantic salmon are reared commercially world-wide. The mass production of cultured fish necessitates the use of large amounts of broodstock. To facilitate production, GnRH implants have been sought to synchronize spawning times of large populations of broodfish. ESC trials and informal trials in Europe and South America have been or are under way. Additionally, three studies involving commercial farms in B.C. and Atlantic Canada are underway. One study is being simultaneously conducted at the University of Victoria and on a commercial farm.

**Objective:**

To induce ovulation in Atlantic salmon and compress the spawning season.

**Design:**

Implanted and control fish were reared in tanks at the University and in pens on the farm. Farm fish were implanted one month in advance of the predicted date of spawn. Fish were transported to the university and implanted 10 days post-transferred.

**Results:**

Implanted fish spawned in advance of control fish by as much as 15 days post implant and 21 days before controls. Males implanted with GnRH pellets gave a higher milt volume than untreated fish.

## Synchronized Spawning of Wild and Captive Broodstock

### Relevance to Managers

By using GnRH implants, gametes can be used as best benefits the breeding program for the stock. Synchronized spawning gives a lever of flexibility that the stock may have lost. Infrastructure and labor can be optimized in the facility. As well, the genetic stock of strains can be preserved.

Fry in the hatchery move through the hatchery system as a uniform group that are faster to hatch and first feeding.

If hatcheries are on surface water, this effect is more pronounced.

In conclusion, GnRH implants are a preferred method of inducing maturation and synchronizing spawning. GnRH implants are under INAD and ESC approval and are safe and easy to use. The use of hypothalamic hormone implants does not interfere with the philosophy of broodstock management.

Powell

# **An Overview of the Captive Broodstock Program in NE Oregon**

William T. Noll

*Oregon Department of Fish and Wildlife, Eastern Oregon University  
1410 L Ave., 211 Inlow Hall, LaGrande, Oregon, 97850*

## **Introduction**

This paper is intended to provide a general overview of the spring chinook captive broodstock program in northeast Oregon. Why a captive broodstock program may be implemented, what activities are entailed in such a program, and how a captive broodstock program may impact day-to-day hatchery activities will be discussed.

## **Evolution of a Captive Broodstock Program**

A captive broodstock program is not one which is undertaken simply out of interest or a desire to be on the cutting edge of fish culture. A traditional salmonid production hatchery probably won't be involved with a captive broodstock program unless it's program incorporates a listed species or, in some cases, a species that is being considered for listing. In NE Oregon the implementation of a captive broodstock program went through an evolutionary process that began with a somewhat steady decline in spring chinook salmon runs over about a 30 year period. During that time, for example, the estimated number of adults returning to spawn in the upper Grande Ronde River dropped from 973 fish in 1968 to three (3) fish in 1994. In 1992 the NMFS listed this stock as threatened. After further review the listing was changed to endangered from mid-1994 to mid-1995; the listing reverted back to threatened in mid-1995. Both the "National Marine Fisheries Service Proposed Recovery Plan" and the "Northwest Power Planning Council's Fish and Wildlife Program" call for the use of captive broodstock programs to prevent stocks from becoming extinct. Therefore in 1995 the Oregon Department of Fish and Wildlife, in conjunction with the NMFS became involved in the NE Oregon spring chinook salmon captive broodstock program in an attempt to delay the possible extinction of three stocks of spring chinook salmon in the Grande Ronde River basin.

## **Program Design**

Realizing that there are many ways to approach captive broodstock technologies, we would like to emphasize that our program is only one of many ways to implement these technologies. Therefore we developed our program objectives based on the endangered status of our spring chinook salmon stocks and the need to maintain the viability and integrity of those stocks. The three

objectives we selected were: 1) to delay the possible extinction of native wild chinook populations in the Lostine and upper Grande Ronde rivers as well as Catherine Creek, 2) to maintain the genetic diversity of indigenous, artificially-propagated chinook populations, and 3) to maintain the genetic diversity in wild chinook populations. While developing the captive broodstock program we also chose to address two related questions: a) How does the performance of fish reared in freshwater as post-smolts compare to that of fish reared in seawater as post-smolts, and b) How does the performance of fish reared under an accelerated growth profile compare to that of fish reared under a (simulated) natural growth profile?

To accomplish the program objectives and address the related questions our program requires the collection of 500 parr from each of three stocks (Catherine Creek and the Lostine and upper Grande Ronde rivers) from throughout their rearing distribution in their natal streams. One-third of the collected fish from each stock are then reared under one of three treatment scenarios: 1) presmolt natural rearing temperatures associated with freshwater post-smolt rearing, 2) presmolt natural rearing temperatures associated with seawater post-smolt rearing, and 3) presmolt elevated rearing temperatures associated with freshwater post-smolt rearing. These fish are reared at hatcheries until they reach maturity and are spawned. The subsequent progeny are reared at a hatchery until they are smolts. These smolts (150,000 from each stock) are then returned to their natal streams. Within three years of their release it is intended for these smolts to result in a return of at least 150 adults to each natal stream.

## **Results and Discussion**

During their captive life stage, and from each stock and its respective treatment scenario, we collect and analyze data on growth, maturity and survival/mortality. Results to date are preliminary since we have not held fish long enough to have completed one complete captive broodstock life cycle. The presented results, therefore, reflect information which we have following almost two years of data collection on 1994 brood year fish. For the 1994 fish we reared fish under only two of the three proposed treatment scenarios: 1) presmolt natural rearing temperatures associated with freshwater post-smolt rearing, and 2) presmolt natural rearing temperatures

associated with seawater post-smolt rearing. No statistical analyses of the data have been completed at this time.

### Growth

For each stock, regardless of whether they were reared in a post-smolt freshwater or seawater environment, the results were very similar (Table 1). Actual growth very closely approximated the predicted growth up to about

eight months following collection. At that point the fish went through smoltification and were moved to their respective rearing sites in freshwater or seawater. From that point onward the actual growth was considerably less than the predicted growth. The actual fork length at age three ranged from 298 mm to 355 mm while the predicted fork length was 480 mm (Table 1). The implications of the smaller-than-predicted size of the captive broodstock is that we may experience maturing females with low fecundity, smaller than desired egg size and low egg viability.

Table 1. Predicted and actual growth of 1994 captive broodstock from Catherine Creek and the Lostine and upper Grande Ronde rivers as measured in mm fork length.

Time of data collection	Predicted Length (mm)	Actual fork lengths in millimeters by stock and growth regime				
		<u>Catherine Creek</u>		<u>Lostine River</u>		<u>Upper Grande Ronde</u>
		Freshwater	Seawater	Freshwater	Seawater	Freshwater
At collection	80	82	82	68	68	90
4 mo.	104	105	109	107	107	104
8 mo.	128	127	127	123	123	125
12 mo.	220	193	193	191	182	175
16 mo.	307	----	----	----	----	----
20 mo.	393	----	----	----	----	----
24 mo.	480	324	355	330	334	298

### Maturity

Table 2 summarizes the 1994 captive broodstock maturity data for each stock at ages two and three, and in freshwater and seawater growth environments. Although we anticipated that 0% and 6% of the females would mature at ages two and three respectively, no females matured at either ages two or three. Therefore the numbers of mature fish listed in Table 2 are all males. The listed percentages in Table 2 are percentages of fish which matured at ages two and three out of the entire number of fish that were collected regardless of gender. If we could assume a 50:50 sex ratio at collection then the percent of males maturing at ages two and three should be double the percentages listed in Table 2 (e.g. of the Catherine Creek stock, 22% of the freshwater reared and 18% of the seawater reared fish matured at age 2). Anticipated

maturity rates for age 2, 3, 4 and 5 males were 2%, 35%, 48% and 15% respectively. Therefore, for all stocks and growth regimes the actual maturity rates for age two males were considerably greater than anticipated. The actual maturity rates for age three males, regardless of stock and growth regimes, more closely approximated the anticipated maturity rates. Once we have spawned all captive broodstock from one year class we will be able to more accurately discuss both numbers and percentages of fish which matured at specific ages. Until that time our data on maturity by sex and at specific ages are only estimates.

## Overview of the Captive Broodstock Program in N.E. Oregon

Table 2. Maturation and mortality of 1994 captive broodstock from Catherine Creek, and the Lostine and upper Grande Ronde rivers.

	Numbers of fish by stock and growth regime									
	<u>Catherine Creek</u>				<u>Lostine River</u>				<u>Upper Grande Ronde</u>	
	Freshwater		Seawater		Freshwater		Seawater		Freshwater	
	No.	%	No.	%	No.	%	No.	%	No.	%
Fish collected	328	----	170	----	328	----	170	----	110	----
Males matured at age 2	36	11%	15	9%	28	9%	5	3%	7	6%
Males matured at age 3	56	17%	36	21%	75	23%	20	12%	13	12%
Mortalities (other than post-spawning)	45	14%	22	13%	83	25%	50	29%	52	47%
Fish remaining	191	58%	97	57%	142	43%	95	56%	38	35%

### Mortality

Mortality data indicates that: there may be a differential mortality rate between stocks for fish that die from causes other than spawning related activities. This data, however, may be misleading as we did not factor out the mortalities which occurred as a result of human error (e.g. fish which jumped out of uncovered tanks etc.). While neither mortality or maturity data is complete at this time it should be noted that for all stocks and all growth regimes we only have between 35% and 58% of the original collected fish still alive. Since all fish that have matured so far were males, and 95+% of the spawned males have died, we have very few males left to spawn with the females which should mature as age four and five adults. The possibility of having almost no fresh males to spawn with the females when they mature places increased significance on the process of cryopreservation of sperm from all spawned males when no ripe females are available.

### Potential Impacts of a Captive Broodstock Program on Day-to-day Fish Culture Operations

If a captive broodstock program is implemented at almost any established hatchery it will have an impact on the day-to-day operations of that hatchery. The magnitude of those impacts will depend, in part, on the specific captive broodstock program design that is chosen, and how closely aligned that design is with current hatchery

practices and the physical capabilities of that facility. If implementing a captive broodstock program such as ours, some of the adjustments which may need to be taken into consideration are compared in Table 3.

Many changes to traditional hatchery practices which are brought about by the implementation of a captive broodstock program are a direct result of the relative importance of each individual fish in the captive broodstock program. For example the current return rate for 150,000 spring chinook smolts released from Lookingglass Hatchery in NE Oregon would be about 0.1%, or 150 adults. Using captive broodstock technologies we are attempting to leverage 500 parr collected from their natal streams into that same number of returning adults (150) within 3 ½ to 5 ½ years following the original collection.

In spite of the changes that have been required at hatchery facilities due to our captive broodstock program we feel that they have been well worth the effort. As stated in our risk assessment summary: *"Given the low escapement levels, we view this approach (Captive Broodstock) as one which will maximize the species reproductive potential as well as individual survival. It also poses the best short-term demographic risk to the remaining wild populations under the present conditions."*

Table 3. A comparison of some traditional fish culture program practices with practices undertaken in a captive broodstock program.

Areas of change	Traditional fish culture program	Captive broodstock program <sup>1/</sup>
Collection	Fish arrive as maturing adults, spawn and die	Fish arrive as parr. Need to transition from a natural diet to an artificial diet and then be reared for from one to three years.
Accountability	A close estimation of pounds and numbers of fish is usually adequate.	Every fish has to be individually accounted for throughout its entire captive life.
Marking	Usually mark large lots of fish (several thousand per lot) with the same fin clip or tag code.	Every fish is given an individual electronic identification number via a PIT tag and then possibly a secondary, visual (VI), individual identification tag.
Tracking	Fish are usually tracked by large lots, often several thousand per lot.	Individual fish are tracked from arrival at the hatchery to their spawning and/or death. Every time a fish is handled or observed it is recorded in a database with associated information.
Handling	Fish are usually crowded, sampled and moved in large groups, often with standard dip nets, crowders, seines etc. Stress to fish is often considered a normal part of handling fish.	Extra precautions are taken to eliminate handling stress (e.g. lighting is adjusted, tanks are covered, special crowders are used, sanctuary dipnets are employed, rearing areas are screened off to reduce outside disturbances etc.).
Spawning	Spawning is usually a matter of selecting random males to spawn with random females as they mature synchronously.	Matrix spawning of individually identified males with specific females of the same stock will be required. If females are not ripe when the males are, semen will be cryopreserved for later use. Females may be spawned with a combination of fresh male semen and cryopreserved semen.

<sup>1/</sup> Activities described may be unique to the NE Oregon captive broodstock program due to the program's research focus and its specific goals, objectives, and the related study questions addressed



# Redfish Lake Sockeye Salmon Captive Broodstock Program, NMFS

Thomas A. Flagg, W. Carlin McAuley, Michael R. Wastel,  
Deborah A. Frost, and Conrad V. W. Mahnken

National Marine Fisheries Service, Northwest Fisheries Science Center, Resource Enhancement and Utilization  
Technology Division, Manchester Marine Experimental Station, PO Box 130, Manchester, WA 98353.

Jeffrey C. Gislason

Bonneville Power Administration, Fish and Wildlife Division - EWN4, P.O. Box 3621, Portland, Oregon 97208

**Abstract**—The National Marine Fisheries Service (NMFS) is maintaining captive broodstocks of Endangered Species Act (ESA)-listed endangered Redfish Lake sockeye salmon (*Oncorhynchus nerka*). Captive broodstock programs are a form of artificial propagation where fish are cultured in captivity for most or all of their life cycle. Implementation and refinement of captive broodstocks for the recovery of Snake River sockeye salmon are identified as priorities in the proposed Recovery Plan for Snake River salmon. It is usual for captive broodstocks to mature at an earlier age and smaller size than wild fish. Furthermore, egg viabilities averaging less than 60% are common. Nevertheless, the relatively high fecundity of anadromous Pacific salmon, coupled with potentially high survival in protective culture, allows captive broodstocks to produce large numbers of adults and juveniles to help "jumpstart" depleted populations. Research using non-endangered Lake Wenatchee (Washington) sockeye salmon ranked husbandry methods producing highest survival to maturity as 1) circular tanks supplied with pathogen-free fresh water; 2) circular tanks supplied with pumped, filtered, and UV-sterilized seawater; and 3) seawater net-pens. Methods 1 and 2 have been employed for Redfish Lake sockeye salmon captive broodstocks since they appear to ensure much higher survival than method 3. The NMFS Redfish Lake sockeye salmon captive broodstocks are complementary to those reared by the Idaho Department of Fish and Game (IDFG) and are intended to reduce the risk of catastrophic loss of this valuable gene pool. Juvenile and adult fish captured, held, and spawned by IDFG are the source of NMFS captive broodstocks. Since 1991, only 15 sockeye salmon adults (zero to eight individuals per year) have returned to Redfish Lake. NMFS has captive broodstocks for 1991-, 1993-, 1994-, and 1996-broods (no females returned in 1992, 1995, and 1997). The fish are reared full term in fresh well water, or from smolt to adult in a pumped, filtered, and UV-sterilized seawater system. Pre-spawning adults, eyed eggs, and juveniles are returned to Idaho to aid recovery efforts. NMFS has spawned ten different groups of Redfish Lake sockeye salmon captive broodstock. Fry to adult survival has ranged from 13-81% and currently averages 50%+. Eyed-egg viability of spawners has averaged about 60%. A total of over 600,000 viable eggs have been produced by the program for use in recovery efforts. This captive broodstock egg production translates to a yearly amplification of 55-240 times the number of eggs taken into protective culture. The relatively high juvenile survival of first and second generation captive broodstock currently being reared should result in an annual production of up to 200,000 eggs. It is virtually certain that without the boost provided by these captive broodstocks, Redfish Lake sockeye salmon would soon be extinct.

## Introduction

The Snake River sockeye salmon (*Oncorhynchus nerka*) are a prime example of a species on the threshold of extinction. The last known remnants of this stock return to Redfish Lake in the Stanley Basin in Idaho. In December 1991, the National Marine Fisheries Service (NMFS) listed Snake River sockeye salmon as endangered under the U.S. Endangered Species Act<sup>1</sup> (ESA) (Waples et al. 1991). Since the listing, only a few sockeye salmon adults (15 total, 0-8 per year) have returned to Redfish Lake. The NMFS is developing a recovery plan for Snake River sockeye salmon (Schmitt et al. 1995). The goal of this recovery plan will be to rebuild listed Snake River sockeye salmon within its

historic range. In the interim, recovery efforts are being coordinated through the Stanley Basin Sockeye Technical Oversight Committee (SBSTOC). Membership on the committee includes representatives from NMFS, the Idaho Department of Fish and Game (IDFG), the Bonneville Power Administration (BPA), the Shoshone-Bannock Tribes, other state and federal agencies, and private groups interested in sockeye salmon restoration in Idaho. On the basis of critically low population numbers, SBSTOC members implemented a captive broodstock project in 1991 as an emergency measure to save Redfish Lake sockeye salmon (Flagg 1993; Johnson 1993; Kline 1993; Spaulding 1993; Flagg and McAuley 1994; Flagg et al. 1994; Teuscher et al. 1994; Flagg et al. 1995; Kline and Younk 1995; Johnson and Pravecsek 1995, 1996; Teuscher et al. 1995; Flagg et al. 1996; Teuscher and Taki 1996; Kline and Lamansky 1997; Pravecsek and Johnson 1997; Taki and Mikkelsen 1997).

Captive broodstock programs are a form of artificial propagation. However, they differ from standard hatchery techniques in one important respect: fish are cultured in

<sup>1</sup> Use of the term "species" in the context of ESA can refer to taxonomic species, subspecies, and distinct population segments. The definition of what constitutes a species under the ESA is addressed by Waples (1991).

captivity for the entire life cycle. Increased survival in protective culture provides the ability for captive broodstocks to rapidly increase effective breeding population size and markedly aid recovery efforts through production of large numbers of juveniles (Flagg and Mahnken 1995). Captive broodstocks should be viewed as a short-term measure to aid in recovery of the gene pool, and not as a substitute for recovering naturally spawning fish to the ecosystem. However, in concert with efforts to correct causes of decline in stocks at risk of extinction, this technology holds promise as a means of accelerating stock recovery by rapidly increasing the abundance of fish available for restocking suitable habitat.

NMFS Northwest Fisheries Science Center (NWFS) is currently maintaining Redfish Lake sockeye salmon captive broodstocks. The NMFS Redfish Lake sockeye salmon captive broodstocks are complementary to those reared by IDFG (Johnson and Pravecek 1995, 1996; Pravecek and Johnson 1997). Prespawning adults, eyed eggs, and juveniles from NMFS broodstocks are provided to Idaho for use in recovery programs for Redfish Lake sockeye salmon. Researchers at the NWFS have also conducted husbandry experiments to refine captive broodstock technology. These studies, using surrogate Lake Wenatchee (Washington) sockeye salmon, were conducted to identify optimal fish culture strategies prior to implementation with Redfish Lake stock. This paper presents an overview of these NMFS captive broodstock projects.

### **I. Captive Broodstock Research Using Non-endangered Lake Wenatchee Sockeye Salmon**

One of the primary goals when maintaining an endangered species in protective culture is ensuring the highest possible survival. At the time of ESA-listing of Redfish Lake sockeye salmon, very little was known regarding methods to ensure survival of these fish in captive culture (Flagg 1993, Flagg and Mahnken 1995). Most past attempts at captive broodstock culture (McAuley 1983; Harrell et al. 1984, 1985, 1987; Peterschmidt 1991; C. Mahnken and T. Flagg, NMFS, unpublished data) indicated that, for Pacific salmon, full-term culture in pathogen-free fresh water generally resulted in higher survival to spawning and higher percentages of viable gametes than culture in seawater. Therefore, full-term freshwater rearing in pathogen-free water was chosen for initiation of endangered species captive broodstocks (C. Mahnken and T. Flagg, NMFS; K. Johnson, IDFG;

recommendations to the Snake River Salmon Recovery Team, 1991).

At the initiation of our studies, it appeared probable that many past husbandry problems in seawater were related to culture in net-pens exposed to near-surface environmental conditions. Several environmental factors critical to survival (e.g., temperature, salinity, and toxic plankton blooms) are more variable at the surface than in the deeper marine waters preferred by most salmonids. In addition, fish held in net-pens are at risk of escape, natural catastrophes, and predation from marine mammals and birds.

In 1992-1995, we conducted studies to determine if land-based facilities supplied with pumped, filtered, and ultraviolet (UV) sterilized seawater could provide the quality environment necessary for full-term protective culture of salmonids in seawater (Flagg 1993, Flagg and McAuley 1994, Flagg et al. 1996, Flagg et al. in prep.). These studies were carried out in seawater at the NMFS Manchester Marine Experimental Station (Manchester, WA) and in freshwater at a Manchester satellite facility located at the University of Washington's Big Beef Creek (BBC) Fisheries Research Station near Seabeck, WA. These studies were conducted with two year-classes (1990- and 1991-brood) of Lake Wenatchee sockeye salmon. Three replicates of about 300 yearling smolts were placed in each of the following environments: 1) 4.1-m diameter circular fiberglass tanks supplied with fresh (10°C) well water at BBC (FWT treatment); 2) 4.1-m diameter circular fiberglass tanks supplied with pumped, filtered, and UV-sterilized seawater at Manchester (SWT treatment); and 3) 4.9-m square seawater net-pens at Manchester (SWP treatment). Water depth in each rearing environment was adjusted to provide about 12 m<sup>3</sup> of fish rearing space.

Survival of 1990-brood Lake Wenatchee sockeye salmon reared for 28 months prior to spawning averaged about 32% for fish held in the FWT treatment; 35% for fish held in the SWT treatment; and 26% for fish held in the SWP treatment (Fig. 1A). There was no significant difference ( $P > 0.05$ ) in survival between the treatments. Survival to spawning for the 1991-brood averaged about 88% for fish held in FWT, 61% for fish held in SWT, and 22% for fish held in SWP (Fig. 1A). For the 1991-brood, there were significant differences ( $P < 0.02$ ) in the percentage of fish remaining in the experiment, with the treatments ranked as FWT > SWT > SWP.

## Redfish Lake Sockeye Salmon Captive Broodstock Program

The high mortality in all groups of the 1990-brood was attributed to severe infections of *Renibacterium salmoninarum*, the causative agent of bacterial kidney disease (BKD). For the 1991-brood, BKD was noted as a severe problem only in fish reared in net-pens. We felt that the 1991-brood data was most indicative of survival under normal rearing conditions. The data confirms that culture in seawater net-pens can result in low survival for sockeye salmon (Fig. 1A). Seawater net-pen culture appears to be a poor choice for sockeye salmon, and most other, captive broodstock programs. However, culture to adulthood in either fresh well water or filtered and UV-sterilized seawater appears to have the potential to provide higher survivals (Fig. 1A) and both strategies appear to be good choices for captive broodstock programs.

For both brood-years, growth differences ( $P < 0.05$ ) were noted, with the treatments ranked  $\text{FWT} > \text{SWT} > \text{SWP}$  for the 1990-brood and  $\text{FWT} > (\text{SWT} = \text{SWP})$  for the 1991-brood. At spawning, 1990-brood Lake Wenatchee sockeye salmon reared in the FWT treatment averaged about 44% larger in weight than fish reared in SWT treatment and 126% larger than fish reared in SWP treatment (Fig. 1B). The 1991-brood Lake Wenatchee sockeye salmon reared in the FWT treatment were about 51% larger than fish reared in the SWT treatment and 92% larger than those reared in the SWP treatment (Fig. 1B). Most 4-year-old spawners from FWT were above an expected 1.5+ kg size range for Columbia River sockeye salmon (Mullan 1986, Burgner 1991), while spawners from SWT and SWP were usually smaller. The cause of these growth differences is unclear. Fish in all treatments received approximately the same pelleted ration on a percent body weight/day basis. In the FWT and SWT treatments, pellets not immediately consumed in the water column were often eaten from the bottom of the tanks by the fish. However, ration falling through the net-pen bottom was lost to the fish and may account for the smaller size of fish from the SWP treatment. This does not explain the size differences between fish reared in freshwater and seawater tanks. It is probable that freshwater vs. seawater growth differences were related to osmoregulatory energy expenditures in seawater and the cyclic nature of seawater temperatures compared to the constant temperature of the fresh well water.

There were no significant differences ( $P > 0.10$ ) in eyed-egg survival for the 1990-brood Lake Wenatchee sockeye salmon. Egg viability for these fish averaged about 50%

from the FWT groups, 42% from the SWT groups, and 45% from the SWP groups (Fig. 1C). Significant differences ( $P < 0.06$ ) in egg viability were noted for 1991-brood spawners between the FWT and SWT and SWP treatments. For the 1991-brood fish, average eyed egg viability from the FWT treatment (67%) was over 50% greater than average viability from SWT (43%) or SWP (40%) (Fig. 1C). All eyed-egg survival rates were much lower than the 80 to 95+% often seen from sockeye salmon adults collected from the wild (Mullan 1986; Flagg et al. 1991; K. Johnson, IDFG, 1800 Trout Road, Eagle, ID. 83616. Pers. commun., December 1997). However, these rates were similar to the 30-60% eyed egg survival documented for many captive broodstocks, including both NMFS and IDFG programs for Redfish Lake sockeye salmon (Schiewe et al. 1997). It is apparent that, at present, fish culturists should expect egg viability rates in the 40-60% range for captively reared sockeye salmon.

## II. Redfish Lake Sockeye Salmon Captive Broodstock Culture

NMFS Redfish Lake sockeye salmon protective culture efforts have focused on maintaining genetic lineages for 1991-, 1993-, 1994-, and 1996-broods (no females returned in 1992, 1995, and 1997). These captive broodstocks include: 1) first and second generation progeny of the one female and three male sockeye salmon that returned to the lake in 1991, 2) first and second generation progeny of the two female and six male sockeye salmon that returned to Redfish Lake in 1993, 3) progeny of outmigrating sockeye salmon captured by IDFG from Redfish Lake Creek in 1991 and spawned in 1993, 4) progeny of residual sockeye salmon captured in Redfish Lake by IDFG and spawned in 1993, 5) first and

Fig. 1A

Flagg et al.

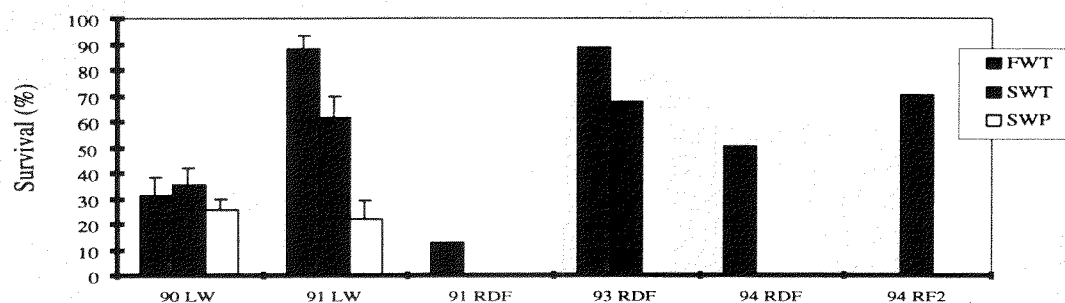


Fig. 1B

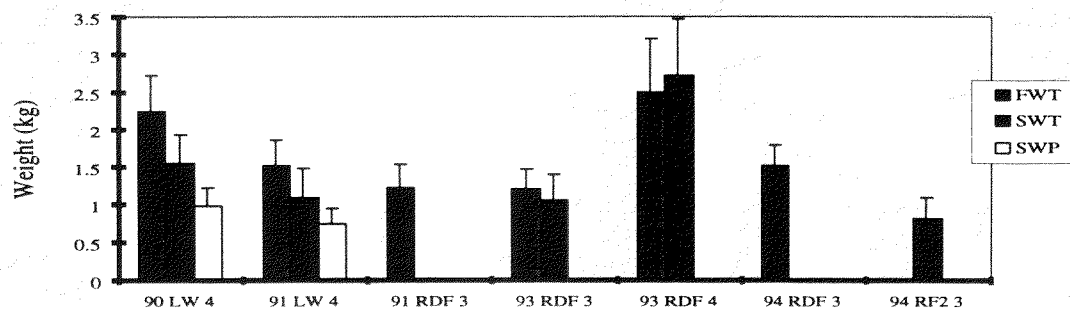


Fig. 1C

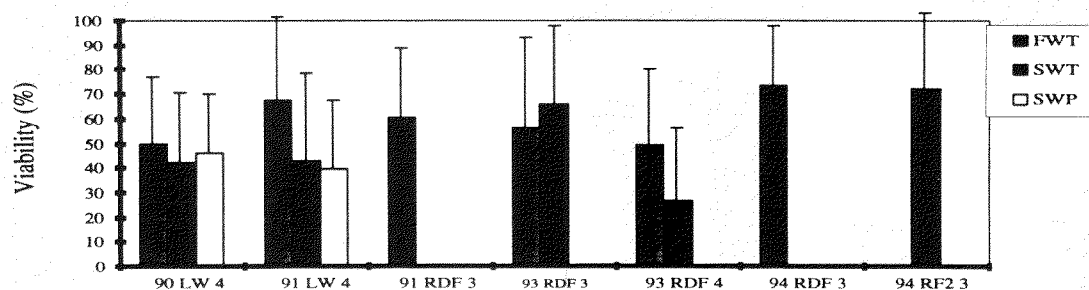


Fig. 1—Prespawning survival (A), female weight (B), and egg viability (C) for 1990-1991 brood Wenatchee (LW) and 1991-1994 brood Redfish Lake 1st (RDF) or 2nd (RF@) generation sockeye salmon captive broodstocks reared in freshwater tanks (FWT), seawater tanks (SWT), or seawater net-pens (SWP). Bars indicate one standard deviation. Note: Redfish groups were not replicated for survival analysis.

second

generation progeny of the one female that returned in 1994, and 6) progeny of the one female sockeye salmon that returned to Redfish Lake in 1996 (Table 1). The NMFS Redfish Lake sockeye salmon captive broodstocks are complementary to those reared by IDFG and are intended to reduce the risk of catastrophic loss of this valuable gene pool. Prespawning adults, eyed eggs, and juveniles are returned to Idaho to aid recovery efforts.

Redfish Lake sockeye salmon captive broodstocks are being reared in 1.8- or 4.1-m diameter tanks using

standard fish culture practices (for an overview of standard methods see Piper et al. 1982, Leitritz and Lewis 1976, Rinne et al. 1986). Fish are fed a commercial diet (e.g., Biodiet<sup>2</sup>) during culture. Therapeutics are

<sup>2</sup> Reference to trade names does not imply endorsement by the National Marine Fisheries Service, NOAA.

# Redfish Lake Sockeye Salmon Captive Broodstock Program

Table 1.— Status of Redfish Lake sockeye salmon captive broodstocks maintained by NMFS, December 1997.

Broodstock year/source <sup>a</sup>	Wild lineage brood year <sup>b</sup>	Initial inventory number	Months in culture	Average survival <sup>c</sup> (%)	Matured			Current inventory number	Egg viability (%)	Adults <sup>d</sup> released (n)	Eyed eggs produced <sup>e</sup> (n)
					female (n)	male (n)	age (years)				
Wild adult returns (F1)											
1991 (eggs)	1987	991	35	13	56	70	3	0	60.4	0	54,000
1993 (eggs)	1989	1,180	38	83	364	523	3	—	53.4	46	270,600
			48	81	26	45	4	0	37.1	0	26,700
1993 (age-3) <sup>f</sup>	1989	151	13	62	16	77	4	0	14.1	0	3,800
1994 (eggs)	1990	461	35	50	65	149	3	0	73.0	36	103,400
1994 (age-3) <sup>f</sup>	1990	304	2	87	—	—	—	265	—	—	—
1996 (eggs)	1992	412	13	60	—	—	—	246	—	—	—
Wild adult residuals (F1)											
1993 (eggs)	1989	58	48	381	1	17	3	—	96.0	0	900
					4	0	4	0	—	4	1,800
Captive-reared adults (F2)											
1993 (eggs)	1989	701	38	70	164	180	3	—	67.3	34	137,800
			48	61	51	33	4	0	28.8	0	29,350
1994 (eggs)	1987	305	35	70	7	36	3	170	72.2	0	6,900
1996 (eggs)	1989	500	13	61	—	—	—	306	—	—	—
1997 (eggs)	1990	296	1	100	—	—	—	296	—	—	—

<sup>a</sup> First generation (F1) captive broodstocks started from eggs from spawners captured from the wild. Second generation (F2) captive broodstocks started from eggs of F1 fish grown to maturity in captivity. F2 broodstocks established to guard against loss of F1 lineages if initial releases to the lakes fail.

<sup>b</sup> Presumed last natural spawning event year is Redfish Lake for age-4 maturing fish.

<sup>c</sup> Captive broodstocks are being held as multiple discrete lots in multiple rearing tanks supplied with either fresh (well) water or filtered and UV-sterilized seawater. Survival percentage is approximate overall average.

<sup>d</sup> Prespawning adults returned to Idaho and released in Redfish, Pettit, or Alturas Lakes.

<sup>e</sup> Most eyed eggs returned to Idaho to aid recovery efforts. Egg total includes estimated viable egg deposition of 17,440 eggs for adults released from F1 1993 brood; 13,400 eggs for F2 1993 brood; and 22,500 eggs from F1 1994 brood.

<sup>f</sup> Age-3 prespawning adults transferred by IDFG to NMFS to alleviate fish rearing space overloads at IDFG Eagle Hatchery.

administered as needed. For instance, diets are modified under FDA New Investigational Animal Drug (INAD) 4333 to contain 0.45% erythromycin and fed at 2% of body weight/day for 28 days on a quarterly basis during rearing as a prophylactic for BKD.

All juvenile fish are reared in fresh 10°C well water at BBC. In most cases, fish are reared as multiple discrete lots in multiple rearing tanks. A majority of the fish are grown full-term to maturity at BBC. However, based on the success of the Lake Wenatchee sockeye salmon captive broodstock experiments described in Section I, the SBSTOC recommended that, in addition to full-term

rearing in freshwater, smolt-to-adult rearing in sterilized seawater be undertaken for select groups of Redfish Lake sockeye salmon. Therefore, at smolting, a portion of the fish may be transferred to tanks supplied with filtered and UV-sterilized seawater at Manchester for culture to adulthood. Juvenile-to-adult rearing density is maintained at under 8 kg/m<sup>3</sup> (0.5 lbs/ft<sup>3</sup>) during most of the culture period; however, fish density may range to 25 kg/m<sup>3</sup> (1.5 lbs/ft<sup>3</sup>) at maturity. Mortalities are examined to determine cause of death. Select mortalities are frozen or preserved as appropriate for genetic or other analyses.

Three brood years of Redfish Lake sockeye salmon captive broodstocks have been reared to adult and spawned (Table 1). Survival from fry ponding to adult spawning for the first captive broodstock (1991-brood) was compromised by BKD and was only about 13% (Fig. 1A and Table 1). Overall survival of subsequent brood years have been substantially higher; averaging in the 50-70% range. These increased survivals have been maintained whether the fish are cultured full term in freshwater or from smolt-to-adult in filtered and UV-sterilized seawater and demonstrate the utility of both rearing approaches in providing adult fish from captive broodstocks to amplify depleted populations. (Fig. 1A and Table 1).

A total of about 98% of 1993-brood fish reared in the FWT treatment and about 41% of fish reared in the SWT treatment matured at age-3 in fall 1996, with the remaining fish maturing at age-4 in fall 1997. Similar maturation disparities were noted for F1 (FWT reared) and F2 (SWT reared) 1994-brood fish (Table 1). It is probable that the lower age-3 maturation ratio for SWT compared to FWT reared fish was due to slower growth in seawater. The availability of culture systems (e.g., FWT and SWT) that alter maturation schedules can be useful in maximizing enhancement opportunities by spreading a broodstocks egg production over a multi year period. The systems can also be used to overlap production from different brood years to help assure availability of males and females for cross-generational mating to promote genetic diversity of broodstocks.

There were slight differences in female spawner weight between groups of 1993-brood reared in the FWT and SWT treatments (Fig. 1B). The size comparison relationship was significant ( $P < 0.05$ ) for age-3 fish but not for age-4 spawning fish (Fig. 1B). As expected, fish maturing at age-4 were significantly ( $P < 0.05$ ) larger in

size than age-3 maturing fish. Eyed egg viability was significantly ( $P < 0.05$ ) greater for 1993-brood fish reared in the SWT treatment compared to the FWT treatment for age-3 fish; however, the reverse was true for age-4 fish (Fig. 1C). There was a large and significant ( $P < 0.05$ ) reduction in egg viability for 1993-brood age-4 compared to age-3 fish for the SWT treatment (27 vs 65%), but not for the FWT treatment. Overall, it appears that egg viability may be slightly improved when fish are reared to mature at age-3 rather than age-4 (Fig. 1C and Table 1). However, more years of data are needed to confirm this relationship.

Overall, more than 600,000 viable eggs have been produced by the NMFS program for use in recovery efforts (Table 1), providing a marked amplification of 55-240 times the eggs taken from wild fish. Additionally, over 120 prespawning adults have been released to Stanley Basin lakes to spawn naturally (Table 1). NMFS adult and eyed egg production complement similar production from IDFG captive broodstock facilities (P. Kline, IDFG, 1800 Trout Road, Eagle, ID. 83616. Pers. commun., December 1997). Together the NMFS and IDFG programs have resulted in near capacity seeding of critical Stanley Basin lake habitats (i.e., Redfish, Pettit, and Alturas Lakes) identified by the SBSTOC as priority enhancement areas. The relatively high juvenile survival of other NMFS broodstock in protective culture (Table 1) should result in production of up to 200,000 eggs yearly for continued enhancement of Redfish Lake sockeye salmon.

### Conclusions

We conclude that captive broodstock technology for salmonids, although still in its initial development stages, is sufficiently advanced to allow carefully planned programs to proceed. In general, fishery managers can anticipate survivals of 50-80% in captive broodstocks if the fish are cultured in water sources low in pathogens. Viability of eggs from captively-reared spawners (especially sockeye salmon) may only range from 30-70% and captively-reared fish may be smaller than wild fish. Nevertheless, captive broodstock programs have the potential to provide the amplification necessary to reduce extinction risk and begin recovery measures for depleted stocks. It is virtually certain that without the boost provided by these captive broodstock projects, Redfish Lake sockeye salmon would soon be extinct.

## Redfish Lake Sockeye Salmon Captive Broodstock Program

### References

- Burgner, R. L. 1991. Life history of sockeye salmon (*Oncorhynchus nerka*). In Groot, C., and L. Margolis (editors), Pacific Salmon Life Histories, p. 1-118. Univ. British Columbia Press, Vancouver, B.C., Canada, 564 p.
- Flagg, T. A. 1993. Redfish Lake sockeye salmon captive broodstock rearing and research, 1991-1992. Report to Bonneville Power Administration, Contract DE-AI79-92BP41841, 16 p. (Available from Northwest Fisheries Science Center, 2725 Montlake Blvd. E., Seattle, WA 98112.)
- Flagg, T. A., and W. C. McAuley. 1994. Redfish Lake sockeye salmon captive broodstock rearing and research, 1993. Report to Bonneville Power Administration, Contract DE-AI79-92BP41841, 99 p. (Available from Northwest Fisheries Science Center, 2725 Montlake Blvd. E., Seattle, WA 98112.)
- Flagg, T. A., K. A. Johnson, and J. C. Gislason. 1994. Redfish Lake sockeye salmon broodstock programs. In Proceedings of the 1993 Alaska Department of Fish and Game Sockeye Culture Workshop. Cooper Landing, Alaska. 10 p. (Available from Northwest Fisheries Science Center, 2725 Montlake Boulevard East, Seattle, WA 98112.)
- Flagg, T. A., and C. V. W. Mahnken (editors). 1995. An assessment of captive broodstock technology for Pacific salmon. Report to Bonneville Power Administration, Contract DE-AI79 93BP55064. (Available from Northwest Fisheries Science Center, 2725 Montlake Blvd. E., Seattle, WA 98112.)
- Flagg, T. A., C. V. W. Mahnken, and K. A. Johnson. 1995. Captive broodstocks for recovery of depleted populations of Pacific salmon. Am. Fish. Soc. Symp. 15:81-90.
- Flagg, T., W. McAuley, M. Wastel, D. Frost, and C. Mahnken. 1996. Redfish Lake sockeye salmon captive broodstock rearing and research, 1994. Report to Bonneville Power Administration, Contract DE-AI79-92BP41841. (Available from Northwest Fisheries Science Center, 2725 Montlake Boulevard East, Seattle, WA 98112.)
- Flagg, T., W. McAuley, M. Wastel, D. Frost, and C. Mahnken. In Prep. Redfish Lake sockeye salmon captive broodstock rearing and research, 1995-1997. Report to Bonneville Power Administration, Contract DE-AI79-92BP41841. (Available from Northwest Fisheries Science Center, 2725 Montlake Boulevard East, Seattle, WA 98112.)
- Flagg, T. A., J. L. Mighell, T. E. Ruehle, L. W. Harrell, and C. V. W. Mahnken. 1991. Cle Elum Lake restoration feasibility study: fish husbandry research, 1989-1991. Report to Bonneville Power Administration, Contract DE-AI79-86BP64840, 52 p. (Available from Northwest Fisheries Science Center, 2725 Montlake Boulevard East, Seattle, WA 98112.)
- Harrell, L. W., T. A. Flagg, and F. W. Waknitz. 1987. Snake River fall chinook salmon broodstock program (1981-1986). Report to Bonneville Power Administration, Contract DE-AI79-83BP39642, 24 p. (Available from Northwest Fisheries Science Center, 2725 Montlake Boulevard East, Seattle, WA 98112.)
- Harrell, L. W., T. A. Flagg, T. M. Scott, and F. W. Waknitz. 1985. Snake River fall chinook salmon broodstock program, 1984. Report to Bonneville Power Administration, Contract DE-AI79-83BP39642, 19 p. (Available from Northwest Fisheries Science Center, 2725 Montlake Boulevard East, Seattle, WA 98112.)
- Harrell, L. W., C. V. W. Mahnken, T. A. Flagg, E. P. Prentice, F. W. Waknitz, J. L. Mighell, and A. J. Novotny. 1984. Status of the National Marine Fisheries Service/U.S. Fish and Wildlife Service Atlantic salmon broodstock program. Report to NMFS/NER, 16 p. (Available from Northwest Fisheries Science Center, 2725 Montlake Boulevard East, Seattle, WA 98112.)
- Johnson, K. A. 1993. Research and recovery of Snake River sockeye salmon, 1991-1992. Report to Bonneville Power Administration, Contract DE-BI79-91BP21065, 38 p. (Available from Idaho Department of Fish and Game, Boise, ID 83707.)
- Johnson, K. A., and J. J. Pravecsek. 1995. Research and recovery of Snake River sockeye salmon, 1993. Report to Bonneville Power Administration, Contract DE-BI79-91BP21065, 37 p. (Available from Idaho Department of Fish and Game, Boise, ID 83707.)

- Johnson, K. A., and J. J. Pravecek. 1996. Research and recovery of Snake River sockeye salmon, 1994-1995. Report to Bonneville Power Administration, Contract DE-BI79-91BP21065, 43 p. (Available from Idaho Department of Fish and Game, Boise, ID 83707.)
- Kline, P. 1994. Research and recovery of Snake River sockeye salmon, 1993. Report to Bonneville Power Administration, Contract DE-BI79-91BP21065, 52 p. (Available from Idaho Department of Fish and Game, Boise, ID 83707.)
- Kline, P., and J. Younk. 1995. Research and recovery of Snake River sockeye salmon, 1994. Report to Bonneville Power Administration, Contract DE-BI79-91BP21065, 46 p. (Available from Idaho Department of Fish and Game, Boise, ID 83707.)
- Kline, P. A., and J. A. Lamansky. 1997. Research and recovery of Snake River sockeye salmon, 1995-1996. Report to Bonneville Power Administration, Contract DE-BI79-91BP21065, 78 p. (Available from Idaho Department of Fish and Game, Boise, ID 83707.)
- McAuley, W. C. 1983. DOMSEA coho broodstock program-update. In T. Noshio (editor), Salmonid broodstock maturation, p. 65-66. Proceedings of the salmonid broodstock maturation workshop. Univ. Washington Sea Grant Pub. WSG-WO 80-1.
- Mullan, J. W. 1986. Determinants of sockeye salmon abundance in the Columbia River, 1880s- 1982: a review and synthesis. U.S. Fish Wildl. Serv. Biol. Rep. 86(12), 136 p.
- Leitritz, E., and R. C. Lewis. 1976. Trout and salmon culture (hatchery methods). Calif. Dep. Fish Game Fish Bull. 164, 197 p.
- Peterschmidt, C. J. 1991. Broodstock rearing and reproductive success of coho salmon (*Oncorhynchus kisutch*). Master's Thesis, Univ. Washington, Seattle, 138 p., plus appendices.
- Piper, R. G., I. B. McElwain, L. E. Orme, J. P. McCraren, L. G. Fowler, and J. R. Leonard. 1982. Fish hatchery management, 517 p. (Available from U.S. Fish and Wildlife Service, Washington, D.C.)
- Pravecek, J. J., and K. A. Johnson. 1997. Research and recovery of Snake River sockeye salmon, 1995-1996. Report to Bonneville Power Administration, Contract DE-BI79-91BP21065, 30 p. (Available from Idaho Department of Fish and Game, Boise, ID 83707.)
- Rinne, J. N., J. E. Johnson, B. L. Jensen, A. W. Ruger, and R. Sorenson. 1986. The role of hatcheries in the management and recovery of threatened and endangered fishes. In R. H. Stroud (editor), Fish culture in fisheries management, p. 271-285. Proceedings of a symposium on the role of fish culture in fisheries management. Am. Fish. Soc., Bethesda, Maryland, 479 p.
- Schiewe, M. H., T. A. Flagg, and B. A. Berejikian. 1997. The use of captive broodstocks for gene conservation of salmon in the western United States. Bull. Natl. Res. Inst. Aquacult., Suppl. 3:29-34.
- Schmitt, R., W. Stelle, Jr., and R. P. Jones. 1995. Proposed Recovery Plan for Snake River Salmon. 347 p., plus appendices. (Available from National Marine Fisheries Service, 525 N.E. Oregon, Suite 500, Portland, OR 97232-2737.)
- Spaulding, S. 1993. Snake River sockeye salmon (*Oncorhynchus nerka*) habitat/limnological research, 1992. Report of research to BPA, Contract DE-BI79-91BP22548, 78 p. (Available from Shoshone-Bannock Tribe, Fort Hall, ID.)
- Taki, D. and A. Mikkelsen. 1997. Snake River sockeye salmon habitat and limnological research, 1996. Report to Bonneville Power Administration, Contract DE-BI79-91BP22548, 97 p. (Available from Bonneville Power Administration, P.O. Box 3621, Portland, OR.)
- Teuscher, D., D. Taki, W. A. Wurtsbaugh, C. Luke, P. Budy, H. P. Gross, and G. Steinhart. 1994. Snake River sockeye salmon habitat and limnological research, 1993. Report to Bonneville Power Administration, Contract DE-BI79-91BP22548, 136 p. (Available from Bonneville Power Administration, P.O. Box 3621, Portland, OR.)
- Teuscher, D., D. Taki, W. A. Wurtsbaugh, C. Luke, P. Budy, and G. Steinhart. 1995. Snake River sockeye salmon habitat and limnological research, 1994. Report to Bonneville Power Administration, Contract DE-



Redfish Lake Sockeye Salmon Captive Broodstock Program

- BI79-91BP22548, 137 p. (Available from Bonneville Power Administration, P.O. Box 3621, Portland, OR.)
- Teuscher, D., and D. Taki. 1996. Snake River sockeye salmon habitat and limnological research, 1995. Report to Bonneville Power Administration, Contract DE-BI79-91BP22548, 85 p. (Available from Bonneville Power Administration, P.O. Box 3621, Portland, OR.)
- Waples, R. S. 1991. Definition of "species" under the Endangered Species Act: application to Pacific salmon. U.S. Dep. Commerce, NOAA Tech. Memo. NMFS F/NWC-194, 29 p.
- Waples, R. S., O. W. Johnson, and R. P. Jones Jr. 1991. Status review for Snake River sockeye salmon. U.S. Dep. Commerce, NOAA Tech. Memo. NMFS F/NWC-195, 23p.



## Dungeness Chinook Freshwater Captive Broodstock Program

Daniel Witczak

*Washington Department of Fish and Wildlife*

Due to the low numbers of adult chinook returning to the Dungeness River, and the potential loss or extinction of the population, a technical committee was formed in December of 1991, and tasked with developing recommendations for rebuilding the Dungeness chinook to a healthy sustainable naturally-spawning population. One of the recommendations from this committee was to use captive broodstock technology as a recovery tool for the Dungeness.

Starting in 1992, known redds were sampled by hydraulic sampling techniques to extract either pre-emergent fry or eyed eggs. Each "family" was reared separately in 4-foot circular tanks with 22 cubic feet of rearing space and 4 gallons-per-minute water now. At approximately 20 fish-per pound, these were tagged using a visual implant tag in the adipose eye tissue, and a coded-wire tag in the nose and adipose fin. Fish collected in excess of program requirements are released back to the river near their original capture point prior to tagging.

Half the program population is transferred to the South Sound net pens for rearing in saltwater. Splitting the production in half helps to ensure against loss of any one or more brood years, should disaster strike. Additionally, it allows us to compare saltwater versus freshwater broodstock performance in chinook.

All brood years are currently being grown to maturity at the Washington Department of Fish and Wildlife Hurd Creek Hatchery. After the fish are tagged, families are combined and transferred to 20-foot diameter circular tanks for grow-out, utilizing 1,250 cubic feet of rearing space, with 100 gallons per minute of water flow. The fish are held in the 20-foot tanks without handling or sampling until late July of the following year, when mature fish are sorted out of the population. Mature fish are transferred to Dungeness Hatchery for spawning. Early rearing of fry is accomplished on surface water at the Dungeness Hatchery. At various stages, different groups of juveniles are transferred to acclimation ponds in the upper watershed. These fish are held for approximately three weeks for acclimation, then allowed to enter the main stem.

Broodstock collection for the brood years sampled so far shown in Table 1.

There have been three successful years of spawning mature freshwater captive brood chinook. The three-year age class in 1995 yielded 42,800 eggs with a green-egg-to-ponding survival rate of 49.2%. The four-year age class in 1996 yielded 1.9 million eggs and green-egg-to-ponding survival of 90.7%. The 1997 spawning of three-, four-, and five-year age classes yielded 2.9 million eggs. We expect survival rates to be consistent with those seen in the 1996 spawn year.

Table 1. Dungeness Chinook Freshwater Captive Broodstock Collection

Brood Year	Number Collected	Families	Number in Fresh Water
1992	3,853	19	3,694
1993	1,520	12	787
1994	3,883	15	1,205
1995	5,846	40	1,189
1996	3,738	46	1,193
1997	2,100 (est.)	12 (est.)	1,200 (est.)

Witczak

# White River Spring Chinook Rebuilding Program Update

Richard M. Johnson

*White River Hatchery, 25305 SE Mud Mt. Road, Enumclaw, WA 98022*

## Introduction

From its source on Mt. Rainier's north face, the glacier-fed White River travels nearly 70 miles westward to its confluence with the Puyallup River at the town of Sumner, WA. The Puyallup River, also of glacial origin, then flows another ten miles to Commencement Bay at Tacoma, WA. Spring chinook once populated several river systems in the South Puget Sound drainage, but are now found only in the White River. Adult returns which once numbered over 5,000 had fallen to less than 30 by the mid 1980's. The declines are thought to be mainly due to fish passage problems associated with the Mud Mountain Dam, operated by the U.S. Army Corps of Engineers, and the White River Hydroelectric Project, operated by Puget Sound Energy (formerly Puget Power). Other impacts on the White River fish runs have included past logging practices, industrial development, flood control measures, and overfishing. White River spring chinook are harvested incidentally throughout their migratory range in British Columbia and Washington with most harvest occurring in Puget Sound sport fisheries.

In the late 1970's, the South Puget Sound Spring Chinook Technical Committee formed to guide the White River spring chinook rebuilding program. The committee is comprised of members from the Muckleshoot Indian Tribe, the Puyallup Tribe, the Nisqually Tribe, the Squaxin Island Tribe, the Washington Department of Fish and Wildlife (WDFW), the U.S. Forest Service (USFS), and the U.S. Fish and Wildlife Service.

## History of Artificial Production (I.)

White River spring chinook have been under some form of artificial production since the early 1970's. In light of the serious habitat and fish passage problems in the White River, WDFW established an off-site egg bank program in 1977 at its Minter Creek Hatchery, near Purdy, WA. Beginning in 1979, juvenile and adult White River spring chinook were held at Hupp Springs, a newly completed satellite facility of Minter Creek Hatchery. All subsequent releases of spring chinook juveniles, until 1990, were limited to Minter Creek, which flows into Carr Inlet. To complement Minter Creek's anadromous returns of White River spring chinook, a saltwater captive brood program

started at the National Marine Fisheries Service (NMFS) net pens at Manchester. This involved the 1977-1986 broods. The captive brood program was moved to the South Sound Net Pen (SSNP) complex, near Olympia, WA, beginning with the 1987 brood. The SSNP is co-managed by WDFW and the Squaxin Island Indian Tribe.

## History of Artificial Production (II.)

The anadromous program expanded in 1989 with the completion of the Muckleshoot Tribe's White River Hatchery. The hatchery was built by Puget Sound Energy (PSE) as part of a settlement agreement between the power company and the Tribe mitigating for the decline of spring chinook and other stocks impacted by hydropower activities in the White River. The White River Hatchery, similar in size to Hupp Springs, doubled the size of the "core" juvenile production program from 350,000 to 700,000. Initially, the White River Hatchery received its eggs and fry from Minter Creek Hatchery or Hupp Springs. In addition, captive brood adults from the SSNP were transferred to both Hupp Springs and White River Hatchery for final holding and spawning. From 1990 through 1997, the SSNP produced 6,409 adult White River spring chinook for the rebuilding program. This resulted in 7.21 million eggs and 4.24 million fry (see Table 1.). A phase out of the captive brood program began in 1995 when the final group of White River spring chinook smolts was taken to the SSNP (1993 brood year). The decision to discontinue smolt transfers to the net pens was based on the increased returns of unmarked chinook to the trap and haul facilities at PSE's diversion dam on the White River at Buckley, WA. (see Table 2.). The final eggtake from captive brood rearing is expected in 1998 (from maturing 5 year-olds). Guidelines for the gradual reduction of the artificial production of White River spring chinook are outlined in the White River Spring Chinook Recovery Plan, developed by the Technical Committee. The Committee meets regularly to discuss the roles of each of the White River spring chinook production facilities and determine the distribution of adult and juvenile spring chinook. It is a primary goal of the recovery program to maintain the genetic integrity of the White River spring chinook population by following the proper spawning protocols and by obtaining positive identification of all brood adults prior to spawning. Coded

wire tags are read before fish are spawned to insure that stray fish are not used in the breeding program.

### **Acclimation Ponds**

AU juveniles produced beyond the needs of the hatcheries are taken to several acclimation ponds in the upper White River watershed for short term rearing, or released directly into the White River system as fry or fingerlings. Over 1.5 million fingerlings have been reared at the acclimation ponds since 1993, when the first pond, at Huckleberry Creek, went into operation. The other two ponds are located on Cripple Creek, a tributary to the West Fork of the White River, and on the Clearwater River. All three acclimation ponds are operated by the Puyallup Tribal Fisheries Department. From 1993 through 1995, fingerlings from the acclimation ponds were trucked down to the White River Hatchery for release below the diversion dam. Since 1996, spring chinook have been released directly from the ponds.

### **Fish Passage**

Two significant fish passage improvements have recently been completed in the White River that will increase the survival of outmigrating juvenile salmonids including spring chinook. Mud Mountain Dam underwent major modifications to strengthen and raise the darn, and to remove valves from the dam's 23 foot diameter tunnel that were causing dangerous conditions for fish moving through the tunnel under high flow conditions. A new inlet arrangement for the tunnel will allow for more natural changes in river flow. Revised operating procedures specify that the reservoir pool be lowered as quickly as possible after storm events to minimize the amount of suspended sediment settling out. In the spring of 1996, a new fish screen and bypass channel was completed to divert fish that enter Puget Sound Energy's diversion flume, at Buckley, WA, back to the river. It replaces an outdated system that allowed a high proportion of fish to enter Lake Tapps from which the only escape was through the penstocks and turbines at the powerhouse.

### **Habitat Restoration**

The U.S. Forest Service has undertaken a number of habitat enhancement projects in the White River drainage in partnership with other agencies and volunteer groups. A large section of the Greenwater River received wood structures to create more pools and retain spawning

gravels. Side channels for juvenile overwintering, and acclimation ponds have also been constructed by the USFS. In the summer of 1997, a group of Boy Scout volunteers, directed by USFS staff, placed log sections in the Cripple Creek acclimation pond to provide cover for the spring chinook that will be held there in 1998. The Scouts also placed spawning gravel in several sections of the creek near the intake and the outlet of the pond.

### **Nutrient Enrichment**

To increase productivity of the major White River tributaries, a nutrient enrichment project was conducted in the fall of 1997. About 1.5 tons of spawned out coho carcasses from the Voights Creek Hatchery were distributed among the Clearwater and Greenwater Rivers, and Huckleberry Creek. The lead organization in the project was the South Puget Sound Salmon Enhancement Group. They were joined by the Washington Department of Fish and Wildlife, the Puyallup and Muckleshoot Indian Tribes, the Weyerhaeuser Co., and student volunteers from the White River High School. Carcass introductions can increase the productivity of a stream reach resulting in a significant increase in the weight of young salmon and a higher number of returning adults, according to research conducted by Bob Bilby, Ph.D., fisheries biologist for Weyerhaeuser.

### **Research**

The first radio-tracking study ever conducted in the Puyallup-White River system is being undertaken by the Puyallup Tribe of Indians Fisheries Department. The goal of the study (now in its second year) is to obtain information concerning entry timing, migration rate, and spawning distribution of returning White River spring chinook. Adults were captured and fitted with stomach implant radio transmitters throughout the migration season (May-Oct.). Fish movement was monitored with the aid of portable receivers and fixed position data logging stations. Tracking was performed by foot, automobile, raft, jet boat, and helicopter depending on stream flows and accessibility. Traditional stream surveys have also been conducted in recent years in the late summer and fall on the main tributaries of the White River. The purpose of these spawning ground surveys is to get live, dead, and redd counts, as well as carcass sampling for coded wire tags, radiotags, GSI and DNA tissues, otoliths, lengths & scales. The Tribes and WDFW each have responsibility for a particular spawning area.

## White River Spring Chinook

### Conclusion

Although the adult population of White River spring chinook has increased dramatically since the mid 1980's, almost 80 % of the stock still exists under some form of artificial production (see Table 3.) The first landmark of success for the rebuilding program will be reached when the target number of 1000 unmarked chinook are collected at the P SE diversion dam (river mile 24.7) and

released above Mud Mountain Dam (river mile 30) to continue on to the spawning ground. The recent corrections of fish passage problems and improvements in habitat conditions may lead to healthier anadromous fish runs in the near future. We are making progress towards the stated goal of rebuilding a harvestable and self-sustaining population of White River spring chinook. To keep on track, we must continue to make good decisions with regard to the use of artificially produced fish to supplement natural production. At the same time, habitat must be protected and restored to meet all of the life history requirements of White River chinook and other salmonids.

Table 1. White River Spring Chinook Captive Brood Summary (South Sound Net Pens).

Year	Adults			Eggs (millions)	Fry (millions)
	F	M	T		
1990	68	13	81	0.16	0.13
1991	664	477	1,141	1.46	0.74
1992	632	523	1,155	1.27	0.73
1993	646	247	903	1.07	0.67
1994	607	572	1,179	1.16	0.80
1995	415	437	852	0.99	0.52
1996	247	364	611	0.49	0.25
1997	352	135	487	0.61	0.40
Total	6409			7.21	4.24

Table 2. Mud Mountain Dam, Fish Haul Report. <sup>1</sup>

Return Year	Chinook	Chinook Jacks	Coho	Coho Jacks
1990	275	0	5,834	6
1991	194	96	4,548	53
1992	406	17	1,264	0
1993	409	28	1,379	3
1994	392	182	6,503	0
1995	605	208	2,733	0
1996	628	95	927	0
1997	402	92	7,216	0

<sup>1</sup> Information provided by the U.S. Army Corps of Engineers.

Table 3. White River Spring Chinook Adults from Artificial Production 1990-1997.

Year	Minter/Hupp (anadromous)	White River (anadromous)	SSNP (captive brood)	Total
1990	234	0	81	315
1991	232	0	1,141	1,373
1992	465	170	1,155	1,790
1993	410	20	903	1,520
1994	316	519	1,179	2,014
1995	565	652	852	2,069
1996	604	924	611	2,139
1997	383	766	487	1,636



# Session IV

## Hatchery Practices

Session Chairs:

Trent Stickell/George Nandor  
(Oregon Department of  
Fish and Wildlife)

# **State-of-the-Art Aquaculture Techniques, Including Air-Cleaning Lines for Waste Removal, Automatic Water Sampling for Nutrients, and Other New Technology for Feed Delivery, Disinfecting Raceways, and Deterring Birds**

Tom Tighe <sup>1</sup>

*Idaho Department of Fish and Game*

Niagara Springs Hatchery is located in southern Idaho near Twin Falls, and is funded by Idaho Power Company (IPC) but operated by Idaho Department of Fish and Game personnel. IPC has a mitigation goal of 400,000 pounds of steelhead smolts annually at Niagara Springs. In 1994, IPC renovated Niagara Springs Hatchery to comply more thoroughly with EPA waste discharge guidelines and also added state-of-the-art culture modifications.

This video presentation will show some of the modifications IPC added to the hatchery to improve fish culture practices. A chain-drive system is used to move

the large feeding bridge over the raceways, while a "bean-bin vibrator" delivers fish feed to a "shaking" fines separator. A water-spray bird deterrent system was installed, along with an air-cleaning system for waste removal from the raceways. Two large settling ponds were added to convert the old "off-line" waste holding system to a constant flow-through "on-line" system. Portable computerized water samplers collect samples 24 hours per day, twice per week for nutrient analysis. A head-screen free headboard system has been installed, and raceways are disinfected with chlorine from the bridge using a spray bar that spans two raceways.

---

<sup>1</sup> Tom elected to only have the abstract of his talk included in the Proceedings.

Tighe

# "Taming the Beast"

## The Road to Fast, Easy, Simple Computerized Feed Programming

Ted R. Calavan

Oregon Department of Fish and Wildlife, Leaburg Hatchery,  
90700 Fish Hatchery Road, Leaburg, OR 97489

*What we were then is... What we are now...*

*But armed with the knowledge and  
Creativity to achieve what we can be...*

*Ted R. Calavan, 1997*

The human being in nature has a unbridled need to improve the environment that they live and work in. Throughout time we have been making advances in technology based on an individuals need to go faster, make work easier, get more accomplished or just to simply make life more manageable. Life at a fish hatchery is no different, the need for constant streamlining is always present with year to year changes in budget and production goals. If we build upon our learned knowledge base and use available technology not as a trend but as a better tool, we can successfully become more efficient and productive.

Today's most technologically based tool is the computer. Today's computers are being used in all walks of life for both work and personal enjoyment. The need for the computer in the work place is constantly increasing as more applications are discovered and others are refined. The use of personal computers in fish hatcheries are no different from any other work place. The applications beyond basic word processing need to be identified or more commonly refined to better meet the needs of the users. One such area of need lies in the number crunching, time consuming task of growth and feed projections.

At Leaburg Trout Hatchery in Oregon we have been using computerized growth programs for a number of years. One such program was labeled "Grow" and was developed by Drew Schaefer of Oregon Department of Fish and Wildlife Nutritional Services back in 1987. This program was written to operate on Lotus Symphony and later versions were adapted to Lotus 1-2-3 and Quattro pro software. The program calculates fish growth based on **Temperature Units required for one inch of growth**. The number of temperature units required per inch of growth varies from stock to stock and possibly year to year. The program uses as default values: 1020 temperature units for Coho salmon, 840 for Chinook, 810 for Steelhead, and 640 for Rainbow Trout. These default

values can be changed if you prefer some other values. I have found the value for trout to be fairly accurate at Leaburg Hatchery. This growth based on temperature units is calculated on a daily basis and the daily change in length is called  $\Delta L$ .  $\Delta L$  fluctuates with changing water temperatures. The average daily temperatures are plugged into the program for the water system it is being used on. A Standard Environmental Temperature (SET) of 59° F has been established for Salmonids. At this temperature, maximum Allowable Growth Rate (AGR) of salmon and trout can be maintained as long as all other environmental parameters are also at optimum. For each degree F above SET, growth rate decreases by 8.25%. Growth rate also decreases as water temperature decreases below SET, and approaches zero as water temperatures decrease below 38° F. The "Grow" program will automatically calculate temperature units,  $\Delta L$ , and fish growth based on water temperatures entered for the stream system. Since most fish hatcheries determine fish growth based on weight gain (#/lb) a condition factor (K factor) is used to convert increases in length ( $\Delta L$ ) to weight (#/lb). An average K factor for salmon is 0.00037 and for trout 0.00045. It is suggested that you use these values until you know the actual values for the fish you are rearing. To then manipulate the growth of the fish on the program you adjust the food conversion and the % AGR to obtain the desired end size for the growth period.

At Leaburg Hatchery we used the Symphony version of "Grow" for several years. I found it to be an effective tool for our purposes but was awkward to use and very slow. At Leaburg Hatchery we have a production of 731,750 rainbow trout reared in 34 ponds with 233 releases at 3.0 f/lb. We feed an average of 300,00 lbs. of feed each year. With this size of production a good computer growth program is a must. After using the "Grow" program for awhile I decided that there must be a way to update the program to a faster and more user friendly windows environment. With the aid of Roger Warren I made contact with Jean-Paul Lagasse, who is the fish nutritionist for O.D.F.W. Jean had been working with the program for some time and had made some improvements to the original version. I proposed what I had in mind and with great enthusiasm Jean went to work on the project. Through meetings with Jean and having many phone conversations, the first new version was sent to me.

## Calavan

The program was written to operate in the Excel Windows 95 environment. The new "Grow" was mouse driven, was easier to look at, and was tremendously faster operating. Since we had just purchased a new pentium computer we were able to reap the benefits of this new technology. With many more phone conversations Jean and I started making improvements to better meet our needs and improve efficiency of the program. With Jeans expertise at the computer and my input from a users point of view, our partnership paid off with more refinements and a second and third version of the program.

Though we are still working on improvements, the program has been in use and has been a great success. Though the program is not fool proof and is intended to serve only as a guide for the fish culturist, it has saved me

countless hours of work and has proven very effective in rearing trout a Leaburg Hatchery. I feel that it will be a real aid to the art of fish culture for both green and seasoned fish culturist alike. I would like to thank Jean-Paul Lagasse for the countless hours he has devoted to a very successful attempt at turning my needs into workable reality and making life out in the field a whole lot easier. Kudos to you!. I hope others will follow suit in continuing the pursuit for technological improvements in all aspects of our job as fish culturist.

For information about the "Grow" program or how to obtain a working copy please contact Jean-Paul Lagasse at 503-657-2000 ext. 238. For personal feedback on the use of the program please feel free to contact Ted Calavan at 541-896-3294.

NOTE: Additional visuals include a copy of the seven day feed print out showing the difference between the 100% AGR and 60% AGR, and an outlined page of raw data from an actual projected growth period.

Computerized Feed Programming

Table 1.-- Leaburg Hatchery Rainbow Trout Growth Projection 10/06/07 to 11/24/97

Pond	Fish/lb Actual Size	Pound Feed Fed	<u>Sampled on 11/25/97</u>		%AGR	+/-	Pounds Gain	Conversion
			Fish/lb Projected size to feed	Pounds				
5	7.8	1427	7.5	1367	95	-.30	1195	1.19
7	9.92	821	10.76	667	60	+.84	791	1.04
8	8.62	1093	8.24	928	60	-.38	728	1.50
9	7.57	1040	7.28	984	65	-.29	770	1.35
11	9.51	819	9.86	651	50	+.35	680	1.20
13	8.83	858	8.55	831	60	-.28	732	1.17
15	7.77	1229	7.56	1181	80	-.21	1017	1.21
17	9.25	841	8.69	713	50	-.56	305	2.76
19	8.7	1128	9.18	1071	85	+.48	696	1.62
21	8.96	1112	9.02	1120	88	+.06	1030	1.08
22	6.79	1140	6.43	1071	65	-.36	679	1.68
23	9.66	1302	9.21	1351	75	-.45	1193	1.09
25	7.55	1112	7.4	1062	68	-.15	932	1.19
27	7.13	1131	6.59	1173	72	-.54	813	1.39
29	7.55	893	8.87	817	60	+1.32	1229	.73
31	9.91	761	10.55	723	60	+.64	793	.96
33	26.95	1958	28.26	1832	120	+1.31	1841	1.06
35	6.80	1343	6.21	1228	70	-.59	990	1.36
37	6.73	1348	6.04	1221	65	-.69	742	1.82
39	4.55	1510	5.49	1470	80	+.94	2212	.68

Calavan

# Use of Electronic Tag Detectors to Detect Coded-Wire Tags in Coho at WDFW Facilities in 1997

Stan Hammer

Washington Department of Fish and Wildlife, Hatcheries Program  
600 Capital Way N., Olympia, WA. 98501-1091

## Introduction

In 1995 legislation was passed ( Chapter 372, Laws of 1995 ) directing the Washington Department of Fish and Wildlife ( WDFW ) to externally mark all coho released from its facilities. This law mandated protection of wild chinook and coho while providing an opportunity for a selective fishery on hatchery chinook and coho identified with an external mark ( Chapter 372, Laws of 1995 ). Since tagged fish were previously identified by the missing adipose fin, new technology was required to separate marked fish containing coded-wire tags (CWTs ) from those that have only a clipped adipose fin. The return of two year old male coho salmon in 1997 from a release of 18.6 million coho earlier in the year precipitated the need to use electronic tag detectors.

WDFW is currently field testing two devices made by Northwest Marine Technology (NMT) allowing detection of CWTs and separation of these two groups of fish. The first device is a hand-held wand detector used primarily by sport and commercial samplers at remote locations to sample small numbers of fish. The wand detector is currently being tested at thirteen hatcheries. The second device, the R-8 detector, is less portable but more efficient and is designed to process large numbers of fish. The R-8 detector is currently being tested at seven facilities.

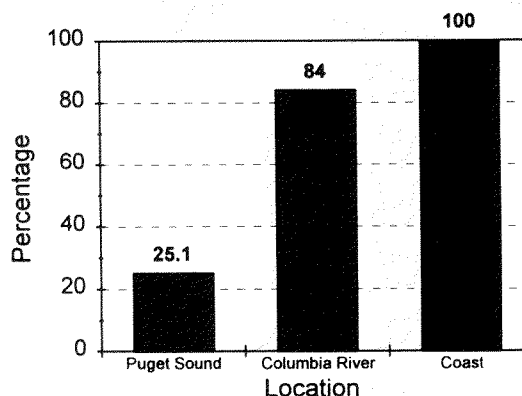
Preliminary evaluation of wands indicate they are reliable but slow, sampling from 100 to 300 fish/hour/person (Bates et al, 1997 ). Field samplers report numerous false readings from background metal objects such as watches, belt buckles, re-bar in concrete, etc. The R-8 detector can sample from 600-800 fish/hour/person (Bates et al, 1997) depending upon adaptations at the hatchery necessary to process fish. Some adjustments may be necessary to use the R-8 for sampling fish to be passed upstream. There are indications that the processing time for sampling both spawned and upstream fish may be increased through the use of these devices. (R. Nicolay, Lewis River Hatchery, WDFW, pers. comm., 1997 ). We expect to electronically sample all returning coho to WDFW facilities in 1998. We will enumerate all fish to develop data indicating hatchery fish/wild fish ratios. This report summarizes the initial effort to electronically recover CWTs from precocious male ( " jack" ) coho returning to WDFW facilities.

## Methods and Materials

### *Criteria for Use of Electronic Tag Detectors*

We expect mass-marked jacks to comprise up to 25% of the total jack returns to Puget Sound facilities and as much as 100% of the total jack returns to WDFW coastal facilities. as high as 22,000 fish ( D. Knutzen, A & D, WDFW, pers. comm., 1997 ), the decision was made to use electronic sampling devices on returning coho jacks to prevent inundating hatchery crews and the tag recovery lab personnel with too many false recoveries. Field trials on the jacks provides WDFW an early evaluation of the electronic tag detectors that will be helpful in planning for the sampling of all returning coho in 1998.

**Mass-marked Coho**  
Percentage by region



**Figure 1. Percentage of mass-marked coho by region.**

### *Distribution of Electronic Tag Detectors*

Wand detectors were issued to the thirteen facilities with the highest expected jack returns ( Table 1 ), and the R-8 detectors were dispensed to the seven facilities expecting tagged fish that had adipose fins (CWT ONLY) returns (Table 1).



*Electronic Sampling of Fish*

Two types of electronic detectors were used to sample the fish both of which were manufactured by Northwest Marine Technology (NMT). The first, and easiest to use, is the hand-held, battery-operated wand detector. It is 40 cm long by 5 cm wide. The wand is operated by switching to the "On" position and rotating the wand pointer over the snout of a fish. The wand either detector costs \$ 5,000. The second device, the R-8, is considerably larger (175 cm long by 37.5 cm wide by 30 cm high and more expensive (\$ 20,000/unit ). The R-8 detects tags by scanning whole fish pushed through the machine, a rounded, rectangular tunnel. Like the wand, the R-8 beeps when a tag is present and remains silent when no tag is present. All jack coho were sampled with either the wand or R-8 detector at the facilities identified in Table 1.

*Enumeration of Marked/Unmarked and Tagged/Untagged Fish*

With the use of electronic tag detectors to separate tagged fish from untagged fish, and the ability to visually separate marked from unmarked fish we can sort the sampled fish into four categories:

- (1) AD +CWT-representing marked and tagged hatchery fish,
- (2) CWT ONLY-representing tagged only wild stock fish,
- (3) AD ONLY-representing ad clipped only hatchery fish, and
- (4) UM-representing unmarked wild stock fish.

Enumerating these groups should provide data on the hatchery fish/wild fish ratio.

**Results****Evaluations of Electronic Tag Detectors***Wand Detectors*

These devices are accurate but slow ( J. Jaquez, Bingham Creek Hatchery, WDFW, pers .comm, 1997 ) depending upon the situation and the experience of the sampler.

**Table 1. Location of Electronic Detectors.**

Facility	Distribution of Electronic Detectors	
	Type of Detector	
	Wand	R-8
Coast:		
Bingham Creek	X	X
Forks Creek	X	X
Humptulips		X
Naselle	X	
Nemah	X	
Satsop Springs	X	
Sol Duc	X	
Puget Sound:		
G. Adams		X
Marblemount		X
Voights Creek		X
Columbia River:		
Elochoman	X	
Fallert Creek	X	
Grays River	X	
Kalama Falls	X	
Lewis River	X	X
North Toutle	X	
Washougal	X	

Because these detectors pick up a lot of background metal readings ( E. Maxwell, Naselle Hatchery, WDFW, pers. comm, 1997), the sampler might consider scanning an area with the wand first to determine if background interference exists before sampling (E. Maxwell, Naselle Hatchery, WDFW, pers. comm. 1997). The wands have "beeped" hooks, watches, ink pens, glasses, etc. Because of the comparative plodding pace needed to scan individual fish, these detectors would not be practical for scanning the large numbers of fish one would expect at most facilities.

*R-8 Detectors*

These detectors are capable of sampling large numbers of fish and are ideally suited for hatchery rack sampling. They can sample up to 800 fish/hour/sampler (G. Britter, Forks Creek Hatchery, WDFW, pers. comm., 1997). The

## Use of Electronic Tag Detectors

R-8s allow sorting of tagged fish from untagged fish into separate totes, significantly reducing time needed to count the marked and unmarked fish. These devices scan dead fish with a very low error rate (Bates et al, 1997). However, we have received conflicting reports regarding the scanning accuracy of live fish (J. Jaquez, Bingham Creek Hatchery, WDFW, pers. comm., 1997). It may be difficult to use the R-8s for upstream sampling of fish because of some unique logistical challenges at some facilities such as lack of access to upstream sampling areas (E. Maxwell, Naselle Hatchery, WDFW, pers. comm., 1997). Use of the "V" tag detector, a portable tag detector used by tagging crews, may be more appropriate for sampling of live fish for passage upstream at some facilities (G. Britter, Forks Creek Hatchery, WDFW, pers. comm., 1997).

### Conclusion

The initial field tests of the electronic detectors have been highly encouraging and as a result of these experiments we expect to accurately sample all returning coho to our facilities providing data on hatchery fish/wild fish composition of runs of coho.

### Acknowledgments

Many people contributed to the completion of this report. The following people provided invaluable assistance:

- \* The following hatchery managers contributed evaluations of the electronic tag detectors: Randy Aho, Dick Aksamit, George Britter, Joel Jaquez, Ed Maxwell, and Rob Nicolay.

- \* Lee Blankenship and Dan Thompson provided technical assistance regarding use of the electronic tagging devices.
- \* Darrell Pruett and Peggy Ushakoff made cartoon slides.
- \* Andy Appleby, Howard Fuss, Mark Kimbel, Hal Michael, and Tim Tynan critiqued this report and provided many helpful suggestions.

### References

- Bates, S., L. Blankenship, R. Boomer, K. Johnson, and R. Olson. 1997. Reliability and feasibility of using electronic detection for recovery of coded wire tags in coho salmon. Pacific Salmon Commission. Ad-hoc Committee Report.
- Chinook and Coho Salmon-External Marking of Hatchery-Produced Fish. 1995. Ch. 372 L.95.( 2SSB 5157 ).

## Hammer

# ENVIRONMENTAL COMPLIANCE AT FISH HATCHERIES

Gary D. Kollman

Senior Environmental Scientist and Toxicologist, Tetra Tech, Inc., 600 University St. Seattle Washington, 98101.

**ABSTRACT**— The day to day operations occurring at fish rearing and research facilities require the storage and handling of hazardous chemicals. These chemicals are used for biological purposes, or for some facilities, hazardous materials are used in exogenous industrial activities (e.g. motor vehicle maintenance) which are vital to the function of the facility. Fish hatcheries must comply with all local, state, and federal environmental laws just like any other industrial facility; although, in some cases agricultural exemptions exist for certain regulations. Environmental and safety and health regulatory compliance for hatcheries can be divided into 14 separate categories. Each category represents a plethora of applicable environmental regulations. These regulations affect such processes as on-site burning, storage and use of hazardous chemicals, construction activities, biological research, pathogen reduction, and grounds maintenance. An external comprehensive multimedia environmental compliance audit can be an excellent and obligatory tool used to identify those areas of fish hatchery operation which may require corrective actions to fix environmental problems. This proactive assessment tool may prevent fiscal losses by prevention of decreased production via hatchery contamination, by eliminating regulatory agency fines, and by decreasing the propensity of work-related accidents. Recent comprehensive environmental audits of fish hatcheries have revealed a large volume of various types of environmental problems. The most common problems are associated with noncompliance with health and safety regulations, incorrect storage and use of hazardous materials, improper wastewater discharge, and deficient storage tanks.

## Assessment Approach and Objective

The primary objective of the environmental compliance assessment process is to identify operations and activities at the Fish Hatchery that may cause adverse impacts to public health and the environment. Specifically, the environmental assessment is intended to evaluate the status of ongoing or unresolved enforcement actions, assess the storage and handling of chemicals and wastes, ensure that personnel maintaining the facility are adequately trained, and evaluate compliance with an Environmental Assessment Manual which is used as a guide to the auditor. The manual organizes environmental regulations into various common topics of compliance.

In accordance with applicable requirements, an assessment is organized under 13 major environmental protocol categories. A supplemental category (Protocol 14 - Environmental Health and Safety Management) is usually added to evaluate compliance with industrial hygiene standards and health and safety procedures. A description of each environmental category and the assessment objectives for each category is presented in Table 1.

To provide a thorough environmental assessment of the Fish Hatchery, the environmental compliance assessment includes a comprehensive records review and facility reconnaissance survey. The following activities are performed during an assessment:

- Review of available facility records, permits, and regulatory correspondence;

- Review of data collected from previous investigations, assessments, monitoring plans, and corrective action programs;
- Review of aerial photographs and maps;
- Inspection/reconnaissance of the fish hatchery to ensure facility compliance with appropriate regulations; and
- Interviews with key, responsible personnel familiar with fish hatchery operations and activities.

Information collected from the facility reconnaissance survey and records review is compared to applicable federal, state, and local regulations to evaluate items of potential environmental concern. The key regulations that serve as references during an assessment include the following:

- The Environmental Assessment and Management (TEAM) Guide, developed by the Department of Defense, 1995;
- Code of Federal Regulations;
- State Administrative Rules; and
- Uniform Fire Code

Non-compliance findings are documented on Environmental Compliance Assessment Individual Finding Sheets. In accordance with applicable requirements, incidents of non-compliance are divided

TABLE 1. — Environmental Compliance Protocols and Objectives

Protocol Number	Protocol Description	Assessment Objectives
1	Air Emissions Management	Evaluate compliance with all applicable regulations associated with air pollution emissions from stationary and mobile sources.
2	Cultural Resources Management	Evaluate compliance with all plans and programs for the protection and management of cultural resources, including historic and prehistoric properties.
3	Hazardous Materials Management	Assess the storage and handling of chemicals and the spill contingency and response requirements related to hazardous materials. Verify that fish hatchery personnel are adequately trained and familiar with hazardous material handling and emergency response procedures.
4	Hazardous Waste Management	Assess the storage and handling of chemicals and the spill contingency and response requirements related to hazardous wastes. Verify that fish hatchery personnel are adequately trained and familiar with hazardous waste handling and emergency response procedures.
5	Natural Resource Management	Evaluate compliance with all permits, plans, and programs for the protection of natural resources and endangered and threatened species.
6	Other Environmental Issues	Evaluate compliance with appropriate regulations concerning the National Environmental Policy Act (NEPA) process, environmental noise, the Installation Restoration Program (IRP), pollution prevention, and program management.
7	Pesticide Management	Assess the storage and handling of pesticides and the spill contingency and response requirements related to pesticides.
8	Petroleum, Oil, and Lubricant Management	Assess the storage, handling, and disposal of petroleum based products and the spill contingency and response requirements related to petroleum products. Verify that fish hatchery personnel are adequately trained and familiar with applicable handling, spill prevention, and emergency response procedures.
9	Solid Waste Management	Evaluate the collection, storage, disposal, and resource recovery of solid wastes generated at the fish hatchery.
10	Storage Tank Management	Review essential regulatory items concerning underground storage tanks (USTs) and aboveground storage tanks (ASTs), including tank emissions, structural concerns, monitoring, and record keeping requirements.
11	Toxic Substances Management	Assess the management of toxic materials, including asbestos, lead based paint, radon, and PCBs.
12	Wastewater Management	Evaluate the regulations, responsibilities, and compliance requirements associated with wastewater and storm water discharge at the fish hatchery.
13	Water Quality Management	Evaluate compliance with all rules, regulations, and requirements associated with quality of the potable water supply system.
14	Environmental Health and Safety Management	Evaluate compliance with industrial hygiene standards and the fish hatchery Health and Safety Program.

## Environmental Compliance at Fish Hatcheries

into two reportable categories: Significant and Major. Significant findings are defined as a situation that requires immediate attention, and poses or is highly likely to pose, a direct and immediate threat to human health, safety, the environment, or the mission. Major findings are defined as situations that require action, but not necessarily immediate action, and may pose a threat to human health, safety, or the environment if uncorrected. Minor findings (mostly

administrative in nature) are noted and verbally transmitted to responsible personnel during the assessment out-brief.

Based on the findings, recommended corrective actions are subsequently developed for each finding. Recommended actions include information on implementability, budget, and responsible party. Exhibit 1 provides a typical example of a Finding Sheet.

### EXHIBIT 1

#### ENVIRONMENTAL ASSESSMENT INDIVIDUAL FINDING SHEET

##### PROTOCOL 3 - HAZARDOUS MATERIALS MANAGEMENT FINDING NO. 1

FACILITY: Fish Hatchery X

DATE: March 18, 1998

BUILDING/LOCATION: Laboratory and Pesticide Storage Room.

FINDING: Major

DESCRIPTION OF FINDING: 1) Flammable materials are being stored next to hydrochloric and acetic acid in the laboratory; and 2) Flammables are being stored next to pesticides in the Pesticide Storage Room. Examples of this finding are shown in Figure X.

CRITERIA: Incompatible materials in storage and incompatible materials in use shall be separated. Reactive compounds should not be stored adjacent to flammable materials.

REGULATORY OR TEAM GUIDELINE CITATION:  
Uniform Fire Code (1994) 8001.9.8

#### CORRECTIVE ACTION(S):

- Immediately segregate the acids and flammable materials.
- Store the flammable materials in the flammable storage cabinet.
- Purchase a corrosive storage cabinet for acid storage.

#### APPROXIMATE COST TO IMPLEMENT CORRECTIVE ACTION(S):

- Up to two person-hours for staff to separate incompatible materials and check Material Safety Data Sheets (MSDSs).
- Up to \$1,000 for purchase of a Corrosive Storage Cabinet.

### Common Findings at Fish Hatcheries

Based on environmental audits at over fifty government fish hatcheries, the following is a list of the most common environmental compliance deficiencies observed at fish hatcheries:

- Improper Open Burning
- Inadequate Underground Storage Tanks
- Minimal Safety and Health Programs
- Improper Storage of Flammable Materials
- Improper Processing of Used Oil
- Improper Transportation of Hazardous Materials
- Improper Designation and Storage of Hazardous Waste
- Unsafe Delivery of Chlorine, Ozone, and/or Formalin
- Improper Storage of Formalin and Chlorine
- Improper Storage of Pesticides
- Improper and/or Non-permitted Wastewater Discharge
- Inadequate Labeling of Hazardous Materials
- Inadequate Training for Staff
- Improper Record keeping for Environmental Records.

Each of these findings may result in staff injury, regulatory agency fines, hatchery contamination, or public relations problems.

### Conclusion

Compliance with applicable environmental and safety and health laws results in fish hatcheries being more productive and safer facilities. Given that the purpose of fish hatcheries and fish research facilities is to increase environmental quality, it is unfortunate and ironic that serious environmental concerns exist at many of these facilities. By performing professional environmental audits, these facilities can take a proactive stance at assuring effective fish production, preventing public relations problems, decreasing liabilities, and assuring a safe workplace for employees. Environmental auditing is a proven, cost-effective means to assure effective hatchery management.

### References

Code of Federal Regulations, Office of the Federal Register National Archives and Records Administration.

Environmental Assessment and Management (TEAM) Guide, developed by the Department of Defense, 1995.

Oregon Administrative Rules.

Washington Administrative Code

Various confidential environmental audit reports prepared by Tetra Tech, Inc. 1997, 1998.

International Fire Code Institute. Uniform Fire Code, 1998.

## Helpful Hints and Tricks of the Trade

John Frost

*Washington Department of Fish and Game, Auburn Maintenance Shop,  
13124 Auburn Black Diamond Road, Auburn WA 98092*

The following are some of the Ideas, Inventions and Short-cuts that are used at various hatcheries throughout the state.

### **"Frogen Valves":** (location Beaver Creek Hatchery)

"Frogen Valves" are a new type of valve that replaces existing "T" handle (or snow valves) used to drain raceway ponds. The "snow valve" handles are screwed into a threaded bracket cemented into the bottom of the pond, the threads wear after time and the bracket has to be removed or replaced which involves cement work and down time. Also the "T" handle has a seal that is exposed to the suction forces in the pond, when you start to release the water the seal is being torn from the bottom plate of the "T" handle and is not able to seal to the cement floor. The Frogen Valve is very easy to use. To release the suction created by the weight of the water at the bottom of the pond I used a cam lever on the lid in conjunction with the handle (operated from the walkway) using a slide hammer effect. By being able to hold the lid open (by using the lock nut on the handle) it also makes it possible to regulate the flow of water to the settling ponds. The seal in the valve is protected from the flow of water by being recessed into the valve and held into place by screws that make the seal replaceable. Installation of the valve is easy!. You use a 1/4" soft rubber seal and use quick bolts to fasten it to the floor over the top of the existing bracket, then bolt the handle to the hand rail (that should be around the ponds).

### **"Walk Thru Pond Ladders"** (Minter Creek Hatchery)

These pond ladders are unique because they allow the worker access to both sides of the pond to work in. They also allow the worker to walk thru the ladders upright hand rails along the center walkway and when the bottom legs of the ladder are folded up, the ladder can be easily moved along the walkway or relocated from pond to pond. These ladders are made from aluminum tubing (1 1/4" sch. 40) and are lightweight enough to be installed by hand.

### **Dump Paddle Alarm Brackets** (throughout the state)

We use a 5 to 10 deg. limit switch with an arm and paddle on these brackets to monitor water flow at various places throughout the hatcheries. The mounting bracket is designed to fit to any size pipe and is attached by using stainless steel bands. By centering the limit switch to the pipe this insures that only the paddle and the arm that attach to the limit switch needs to be sized to the pipe diameter instead of fabricating a specific size bracket for each size of pipe.

### **Sluice Gate** (Soos Creek Hatchery)

An age old problem of removal of rocks and gravel that flow down stream and build up in front of dam-boards and pump stations. The "Sluice Gate" allows water to move under the gate at a regulated flow and allows the sand and gravel to continue down stream while keeping the required amount of water to run the pumps or water systems. The "Sluice Gate" has a 2 ton hoist on the top that can open or close the slide door at the bottom of the gate. When the gate is open it creates a current of water thru the gate and takes the gravel and sand down river and out of the area of the intake. This is very cost effective when the alternative is to dredge using heavy equipment and having to get hydraulic permits.

### **Stop Log Hoist** (Tokul Creek Hatchery)

Removing stop logs can be hard on the back! Here is a 500 lb. Hoist that does the job. The only thing that you have to do is put a bar thru the stop log so the hoist can lift from both sides of the log and you can let the hoist do the lifting.

### **Bird Predation** (Tokul Creek Hatchery)

We have two types of Bird Predation. One type is for the raceways (as shown) using the counterweights in the center of the poles and the other type used for round ponds (20 foot dia.). The round pond structures are made of aluminum and have a 16' wedged shape sliding door for access. Its covered with 4" square netting that is cut to size and is held in place using tie strips along the aluminum and hooks around the cement edge of the pond.



Frost

**Settling Tank (Soos Creek Hatchery)**

During the storm last winter, our settling area for our domestic water went down hill, literally!. We installed an intake to restore water but the intake is at ground level, instead of getting permits and heavy equipment to put in

a new settling pond on the hill, we made a settling tank that allowed us to build the tank in the shop, install it at the hatchery (using a forklift) and to clean the system I installed a 4" blow-off valve. The system is still in the testing stage but seems to be working well.

# Investigation of Rearing & Release Strategies Affecting Adult Production of Spring Chinook Salmon

Douglas E. Olson

*U.S. Fish and Wildlife Service, Columbia River Fisheries Program Office  
9317 Highway 99, Suite I, Vancouver, Washington*

**Abstract.**—For over 15 years, all spring chinook salmon released from Warm Springs National Fish Hatchery have been externally marked to identify them as hatchery fish. This marking program has made it possible to study rearing and release strategies at the hatchery. Our goal in investigating the various release groups is to determine which of several treatments maximize adult yield while minimizing impact upon wild fish populations. In this paper, a number of fish culture questions will be addressed. Can diet and medication affect prevalence of bacterial kidney disease and adult yield? Do fish released in the fall contribute to adult production? Is there differential survival based on size at release? What rearing density will maximize adult yield? Does fin clipping affect returns? Are we still dazed by diet, densities and disease?

## Introduction

The study site is Warm Springs National Fish Hatchery (NFH). Rearing units consist of 2 adult holding ponds, 3 catch ponds, 30 Burrows ponds, and 20 starter tanks. The hatchery is located at River Mile 8 on the Warm Springs River, within the Warm Springs Indian Reservation of Oregon. The Warm Springs River is in the Deschutes River subbasin of the Columbia River. The Deschutes River flows into the Columbia River upstream of Bonneville and The Dalles Dams, 205 miles upstream from the Pacific Ocean. A detailed description of spring chinook salmon and the Deschutes River subbasin can be found in Lindsay et al. (1989) -and- Oregon Department of Fish & Wildlife and Confederated Tribes of the Warm Springs Reservation of Oregon (1990).

Since the start of production in brood year (BY) 1978, spring chinook released from the hatchery have been externally marked to identify them as hatchery fish. This marking program has made it possible to study rearing and release strategies at the hatchery to maximize adult yield while minimizing impact upon wild fish populations (Olson et al. 1995). The external marks applied include ventral and adipose fin clips. The use of an internal coded-wire tag was associated with the adipose fin clip (AdCWT) on spring chinook. Since brood year 1982 all spring chinook production was externally marked and since brood year 1990 was 100% AdCWT.

An osteo-mark was another internal mark used for hatchery evaluation. Oxytetracycline (OTC) in the feed produces white bands "rings" on bony structures in fish (Hendricks et al. 1991). These rings are seen when the bony structure is dissected and viewed under ultraviolet light.

## *Does fin clipping affect returns?*

For 3 brood years, we applied ventral fin clips or AdCWT to juvenile fish in order to evaluate the effect of the two marks on survival to adult. It appears that when comparing ventral fin clips to AdCWT fish, ventral fin clips were not detrimental to survival as previously suspected at Warm Springs NFH (Figure 1). A final report on this marking study is being developed.

## *Hatchery spring chinook rearing and release strategies*

Releases from the hatchery were typically split into fall (subyearling) and spring (yearling) releases. The fall release ranged from < 10% to > 50% of the total production (Figure 2). The fall / spring split release was initiated after observing that the first year's production (BY78) had a bimodal length frequency distribution in the fall, with the larger fish appearing to show signs of smolting (silvery and loss of parr marks). Higher than normal mortality of these larger fish was also observed when they were held overwinter until the following spring.

In September, the large fish were separated by use of a grading device in a fish loading pump, or starting in 1988, passive in-pond graders. The larger fish were released in early October at 9 to 10 per lb. (>140mm @22 per kg). Smaller fish were reared overwinter and released in April at 15 to 20 per lb. (22 per kg).

Oxytetracycline (OTC) was fed to the larger fall release group until brood year 1985. Starting with the 1987 brood, the smaller fish released in spring received OTC. At the hatchery, a section of vertebrae from the caudal peduncle was dissected from adult fish and examined under an ultraviolet lighted microscope for presence/absence of the OTC "ring".

*Do fall releases contribute to adult returns?*

The fish released in the fall do survive and contribute to adult returns. In some years, the larger fall released fish returned to the hatchery at a higher rate (Figure 3), but the larger fish released in the fall also produced a higher percentage of age 3 jacks relative to the age 4 and 5 adult return (Cates 1992).

*Will the larger fish graded in the fall survive if reared overwinter in the hatchery and released the following spring?*

Looking at percent adult recoveries, it appears that the larger fish do survive if held overwinter at the hatchery but at a lower rate than the smaller fish (Figure 4).

*Is the fall/spring pond splitting strategy better than the standard spring yearling release?*

The strategies investigated include the fall / spring split-graded (as previously described) and the fall/spring split-volitional release groups. The partial volitional release typically started 2 to 3 weeks before the forced release date in the fall and spring. Instead of grading, the fish exited the hatchery on their own volition for about 3 to 4 weeks in both the fall (October/November) and following spring (March/April). Approximately 10% of the fish were estimated to exit during the fall period. Past records indicate that a mixture of sizes exit the hatchery from this fall volitional release. To release the fish from the pond, the exit screens were pulled in front of the dam boards (current practice is to place a stand pipe in the drain outlet). The remaining fish were reared overwinter and allowed to exit volitionally from late-March through April. Some fish were also reared overwinter until the yearling spring release period with no fall emigration. This was the standard spring yearling release group.

Looking at percent adult returns, there appears to be mixed results between the spring yearling and fall/spring graded release strategy. The fall/spring volitional release appears to be the best strategy (Figure 5).

*Can rearing environment affect adult returns?*

Starting with brood year 1989 an alternative rearing environment was tested using the adult brood holding ponds. There are two brood holding ponds, approximately 50X26 foot oval shaped with 6 foot water

depth. The raceways are modified Burrow's ponds (30 @ 75X16 foot rectangles with 1.7 foot water depth) and are where juvenile production typically occurs. After spawning adult fish in the fall, the brood ponds were cleaned and approximately 16,000 juvenile fish were transferred to each of the two ponds at a density of around 0.3 lbs/cu ft at release. The burrows ponds received from 26,000 to 52,000 fish per pond or about 1.8 lbs/cu ft for a pond of 39,000 fish at release.

Looking at 3 brood years and percent adult recovery, the fish which overwintered in the adult holding ponds at low densities survived substantially higher compared to fish reared in the typical raceway environment (Figure 6).

*Does rearing density affect adult yield?*

The hatchery has also looked at reduced overwinter rearing densities at 20,000 to 30,000 per pond compared to 50,000 to 60,000 per pond after the fall graded release. Prior to brood year 1987, it was standard practice to grade out the larger fish and release them in the fall, then combine the ponds of remaining smaller fish. Approximately 60,000 per pond of the smaller fish were reared overwinter until the spring release = High Overwinter Rearing Density. Are there any adult survival benefits from not combining the ponds of smaller fish after the fall release? Approximately 30,000 per pond of the smaller fish were reared overwinter until the spring release = Low Overwinter Rearing Density.

For the 1987 and '88 brood years, more adults were recovered from the low overwinter density groups (Figure 7). In Brood year 1989 both groups had poor survival.

To specifically address the question of rearing density and the effect upon adult survival, a rearing density study was developed, modeled after Joe Banks study with spring chinook at Carson NFH (Banks 1994).

For 3 brood years, 3 rearing densities were investigated at 26K, 39K, and 52K per pond -or- 1.2, 1.8, and 2.4 lb/cu ft at release, respectively. The fish received a spring yearling release (no fall/spring split) with 2 ponds per treatment per year with a unique coded-wire tag for each pond.

We have preliminary data on returns to the hatchery for brood year 1992. Brood years 1990 and 1991 survived poorly regardless of rearing density, in part from on-

## Investigation of Rearing & Release Strategies

hatchery disease and otter predation as well as poor off-station survival.

What we see is that percent survival and adult yield per pond was higher for the lower density groups (Figure 8). Again, this is preliminary data but combined with the other information at the hatchery on overwinter densities, density does affect survival to adult, with lower densities producing higher survival rates and number of adults.

### Fish Health Applications

*What about diet and bacterial kidney disease (BKD) of juvenile fish affecting adult survival?*

Two diets were compared, Abernathy Dry and BioMoist. The study was set up by fish health specialists assuming that dry diets enhance the prevalence of BKD. The fish were also marked so adult survival could be determined from the two diets.

Mixed results were observed on the effect of BKD prevalence in the juvenile fish fed the two diets (Mavis Shaw, Warm Springs NFH, February 11, 1997 correspondence). Juvenile fish fed the dry diet had a high prevalence of BKD the first year studied. The following year both diet groups had high BKD prevalence in the juvenile fish and in brood year 1989 the dry diet fed fish had lower BKD levels (Figure 9). Please note that a portion of the dry feed diet was determined to be of poor quality in brood year 1988 which may have also influenced the results.

*What about survival to adult?*

Generally speaking, in two out of three years studied, juvenile fish fed the Abernathy Dry Diet had lower survival to adult, brood year 1989 the exception (Figure 9).

*What is the effect of feeding Erythromycin to juvenile salmon for control of BKD at Warm Springs NFH?*

This study was undertaken under Investigative New Animal Drug 4333. Spring chinook salmon reared at Warm Springs NFH, as well as wild juveniles and adults in the Warm Springs River itself, are infected to varying extent with *R. salmoninarum*. The objectives of the study are to determine the potential benefits of oral erythromycin treatment on the survival of juveniles in the hatchery, the levels of soluble antigen produced by *R.*

*salmoninarum* in juveniles in the hatchery as an indirect measure of the level of infection, and survival to adult. The Lower Columbia River Fish Health Center of the USFWS is the lead investigator. The study was initiated in brood year 1993 and continues to the present.

Drug concentrations in the diet and feeding regimes provided a daily dosage of 100 mg/kg body weight. Control and treatment fish were fed three times daily. Feeding rates were calculated based on feeding 2% body weight of fish per day. The drug was erythromycin thiocyanate (Aquamycin 100). Erythromycin therapy was administered for 21 days for each of two treatments in spring and summer. Juvenile fish health sampling and pond mortality data was also collected.

We have hatchery recovery data that so far includes four year old adult returns from fall 1997 for brood year 1993. We see substantial difference in survival to adult between the control and treatment groups. The groups from the medicated feed survived better than the standard spring yearling release and fall/spring split volitional release groups (Figure 10). Again this is preliminary data. Along with juvenile and adult fish health information, we will be looking at these returns for the next 5 plus years.

*Do progeny segregated by ELISA survive?*

Another on-going Fish Health practice at Warm Springs NFH is Enzyme-Linked Immunosorbent Assay (ELISA) based segregation of eggs and juvenile fish. This practice started with brood year 1984. Adults were also injected with Erythromycin. At time of spawning, adults were sampled and eggs segregated based on the ELISA optical density (O.D.) measurement. Eggs have been culled from females with ELISA O.D. > 0.5. Eggs from females with O.D. < 0.5 were kept for production and segregated into two or more groups. This segregation occurred through the juvenile rearing phase. The juvenile fish were differentially marked to identify them at adult recovery.

So far, only brood year 1993 has shown the expected response, lower BKD ELISA values : higher survival to adult. This relationship was not evident in all other brood years (Figure 11). Please note that higher ELISA groups often were reared at lower densities, which influences survival. Also, the hatchery has an open water supply and resident Warm Springs River fish have BKD (reservoir of infection).

The main point is that ELISA groups with O.D. < 0.5 can survive and contribute to adult returns. But we also need

to look at the data further - Has the proportion of low and high groups changed over time? Do low and high groups that survive produce the next generation of low and high groups? Do ELISA groups with O.D. > 0.5 survive? What is the on-hatchery mortality of the different ELISA groups? Any relationship of juvenile BKD incidence and adult survival?

#### **Future Rearing and Release Strategies at Warm Springs NFH**

*Rearing densities <26,000 per pond (< 1.2 lb/cu ft at release)*

We have seen benefits from reduced rearing densities, but how low can you go and still maximize adult yield? We may want to investigate rearing densities of 19,500 and 13,000 per pond. A good time to experiment with these low rearing densities may be during low adult return years.

#### *Alternative rearing environment*

The hatchery has recently installed 20% shade cloth and sprinklers over the ponds for rearing during the summer and fall months. From what was presented on the NATURES work, we may want to try some alternative rearing environments as well. What about large root wads for in-pond diversity? Increase percent shade >50%?. Could a different pond design be installed?

#### *Improve water quality during the rearing phase*

Water temperature and rearing conditions at the hatchery are less than ideal for raising salmon because the rearing ponds are dependent upon untreated river water. Daily maximum summer water temperatures often hit 20° C. The use of untreated river water for juvenile rearing also creates fish health problems in the hatchery. We are currently developing a plan for disinfection and summer cooling using reuse and ozone treatment.

#### *Diet*

We need to continue application and investigation of the medicated feed program. We could also possibly develop alternative programmed feed schedules for targeting different sizes at release. Are on-demand feeders available and functional for semi-moist diets?

#### *Release Strategy*

We need to be cognizant of the effect of our hatchery releases on wild fish production, particularly our fall releases which overwinter in freshwater before entering the ocean. Can we develop a release strategy which maximizes adult yield and minimizes impact on wild fish? What about volitional releases only during the spring period as yearlings? How can we maximize survival and minimize number of on-station releases?

#### **Conclusion**

*Are we still dazed by diet, densities and disease?*

Examining on-hatchery performance and off-station survival of juvenile fish will only tell part of the story. To determine performance of our rearing and release strategies, we need to examine adult yield.

#### **Acknowledgments**

I wish to recognize contributions from USFWS staff at Warm Springs National Fish Hatchery, Lower Columbia River Fish Health Center, Columbia River Fisheries Program Office, and staff with the Confederated Tribes of the Warm Springs Reservation of Oregon. I wish to specifically acknowledge help from members of the 1997 hatchery evaluation team, Mike Paiya, Mavis Shaw, Randy Boise, Steve Turner, Theresa Kerr, Susan Gutenberger, Colleen Fagan, and our data manager, Steve Pastor. Brian Cates was responsible for Warm Springs hatchery evaluation prior to 1990 (Cates 1992). And without the help of our marking crew (Skip Walch, Steve Olhausen, Dan Magnuson, Pat Kemper and Chuck Fuller), these evaluations would not be possible. Although many people have contributed to this paper, information and views presented in this report are my own and do not necessarily agree with all parties or policies.

#### **References**

- Banks, J.L. 1994. Raceway density and water flow as factors affecting spring chinook salmon (*Oncorhynchus tshawytscha*) during rearing and after release. *Aquaculture* 119:201-217.
- Cates, B.C. 1992. Warm Springs National Fish Hatchery evaluation and anadromous fish study on the Warm Springs Indian Reservation, 1975-1989. Progress report by the U.S. Fish and Wildlife Service, Vancouver, Washington.

### Investigation of Rearing & Release Strategies

Hendricks, M.L., T.R. Bender, and V.A. Mudrak. 1991. Multiple marking of American Shad otoliths with tetracycline antibiotics. *North American Journal of Fisheries Management* 11:212-219.

Lindsay, R.B., B.C. Jonasson, R.K. Schroeder, and B.C. Cates. 1989. Spring chinook salmon in the Deschutes River, Oregon. Oregon Department of Fish and Wildlife Information Report 89-4, Portland.

Olson, D.E. and B.C. Cates and D.H. Diggs. 1995. Use of a National Fish Hatchery to complement wild salmon and steelhead production in an Oregon stream. *American Fisheries Symposium* 15:317-328.

Oregon Department of Fish & Wildlife and Confederated Tribes of the Warm Springs Reservation. 1990. Deschutes River subbasin salmon and steelhead production plan. Report to the Northwest Power Planning Council, Portland, Oregon.

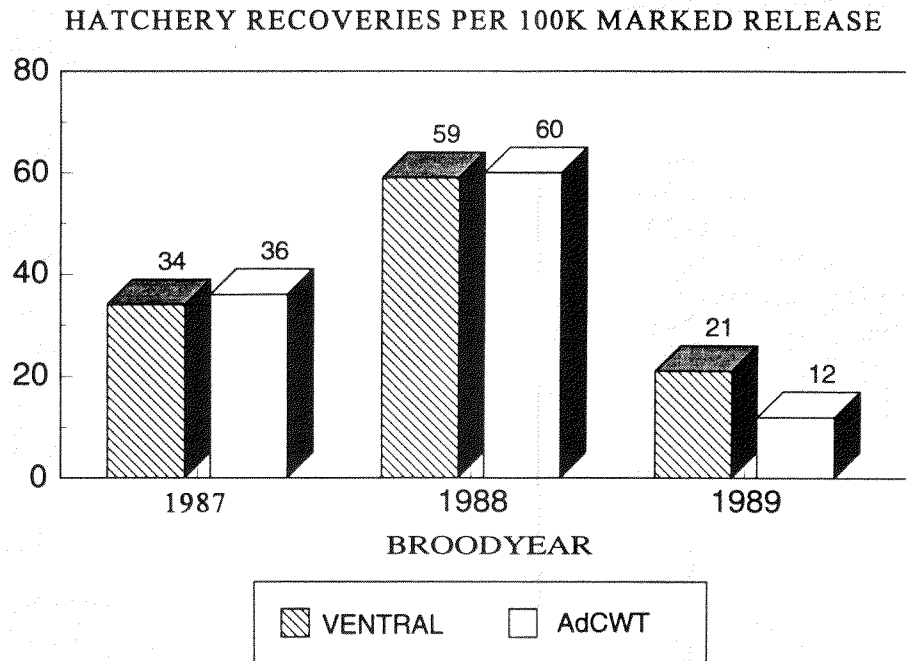


Figure 1. Ventral fin clip vs. Adipose fin clip/Coded-wire tag survival

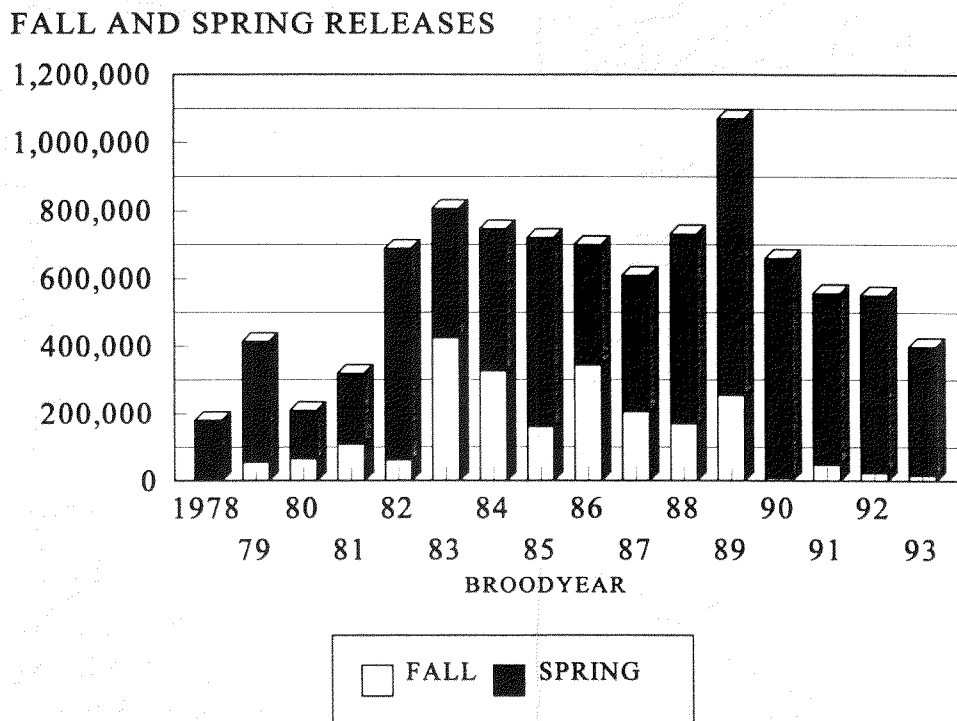


Figure 2. Spring chinook salmon production releases.

RELEASE TO RECOVERY (%)

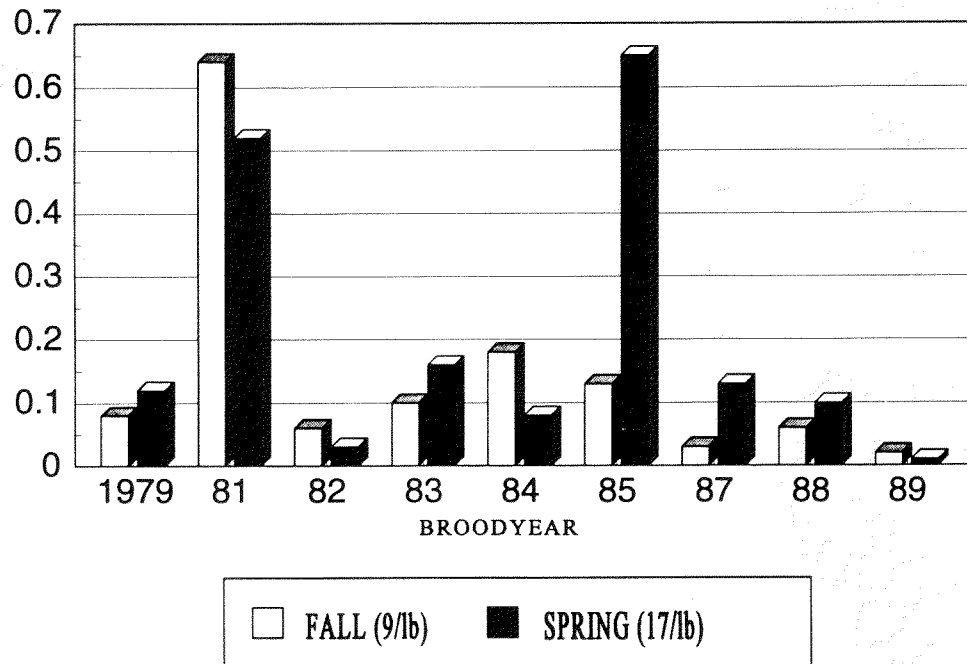


Figure 3. Graded Fall and spring release survival.

RELEASE TO RECOVERY (%)

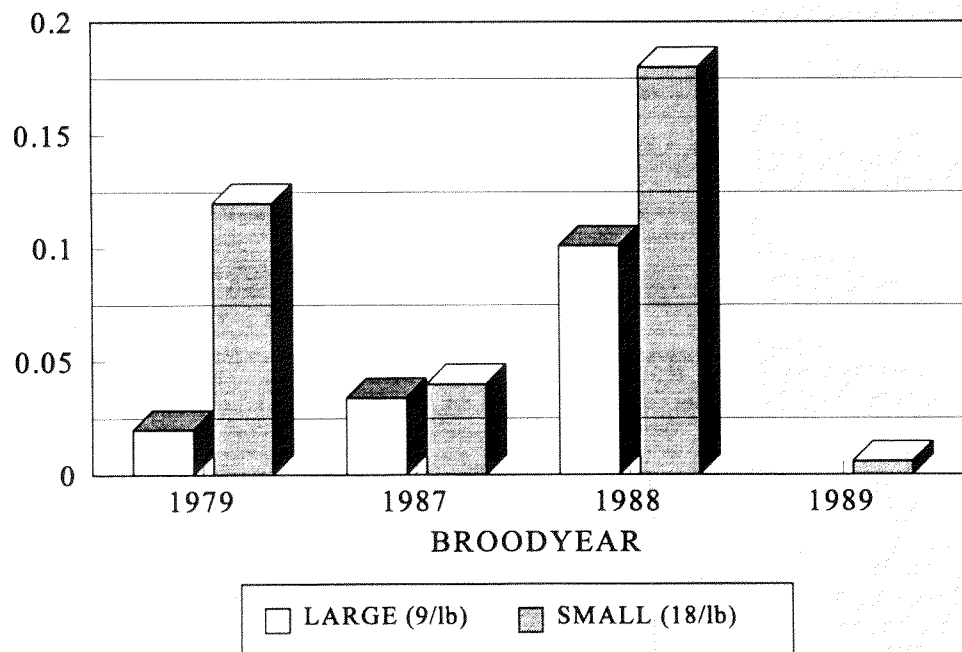


Figure 4. Spring release and survival of large and small fish.



## RELEASE TO RECOVERY (%)

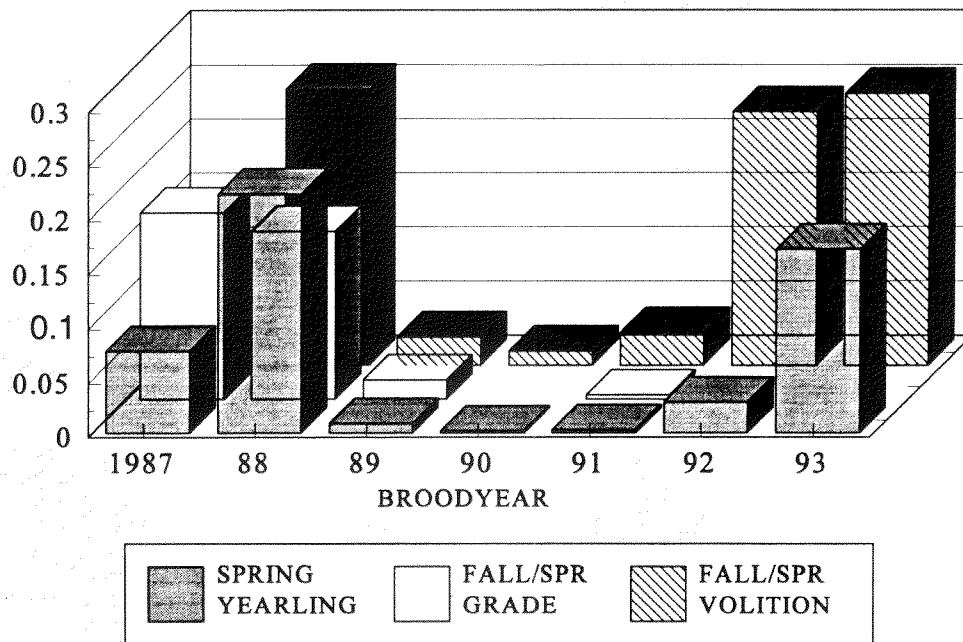


Figure 5. Adult yield of 3 different pond release strategies.

## RELEASE TO RECOVERY (%)

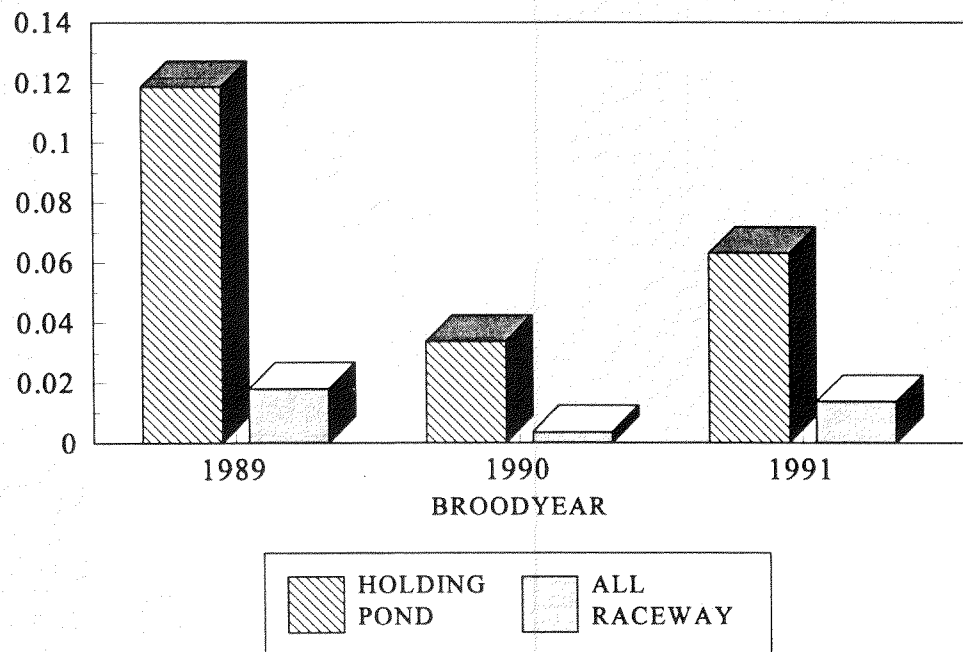


Figure 6. Survival of holding pond overwinter rearing.

# Investigation of Rearing & Release Strategies

## RELEASE TO RECOVERY (%)

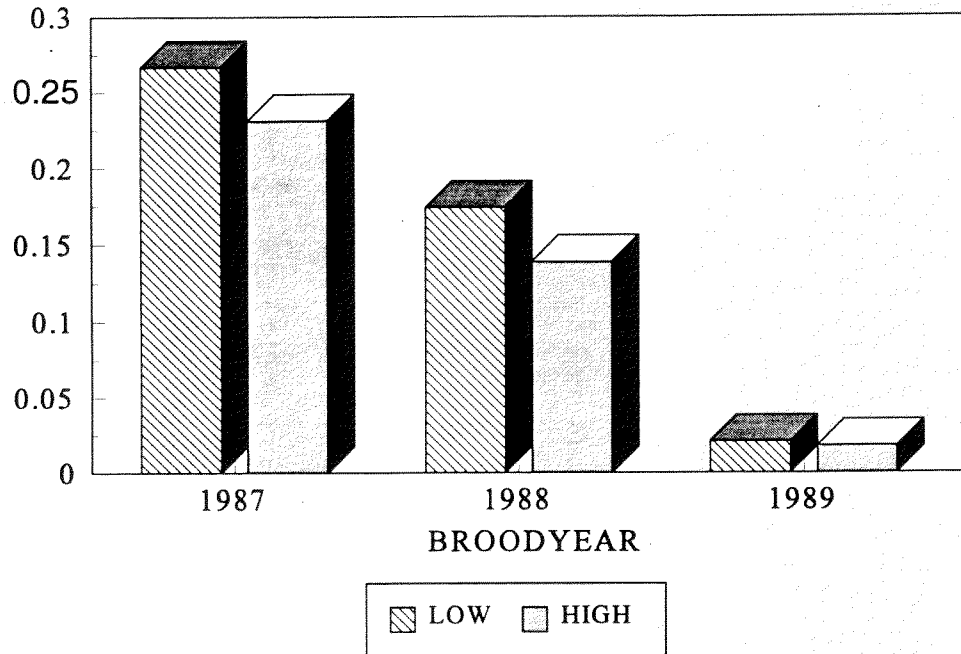
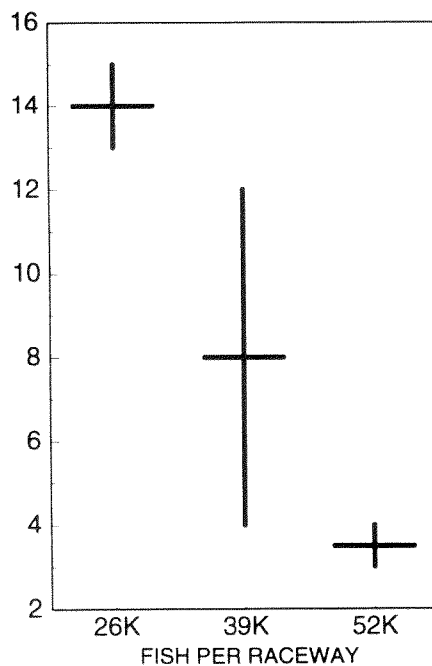


Figure 7. Overwinter rearing and survival at 20K-30K fish/pond (low and 50K-60K fish/pond (high).

## RECOVERY PER POND



## RELEASE TO RECOVERY (%)

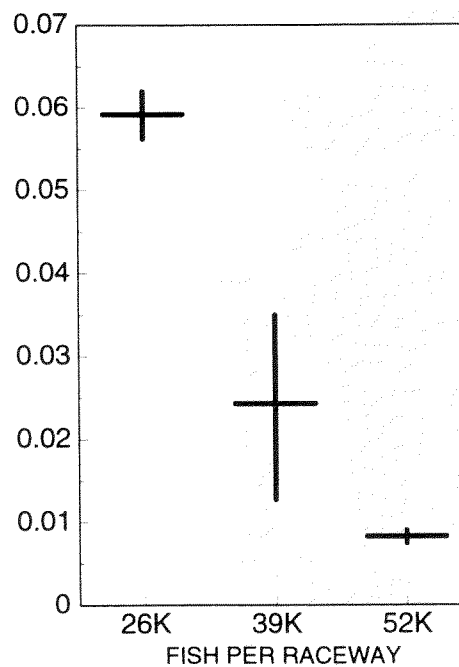


Figure 8. Rearing density study hatchery returns, brood year 1992.

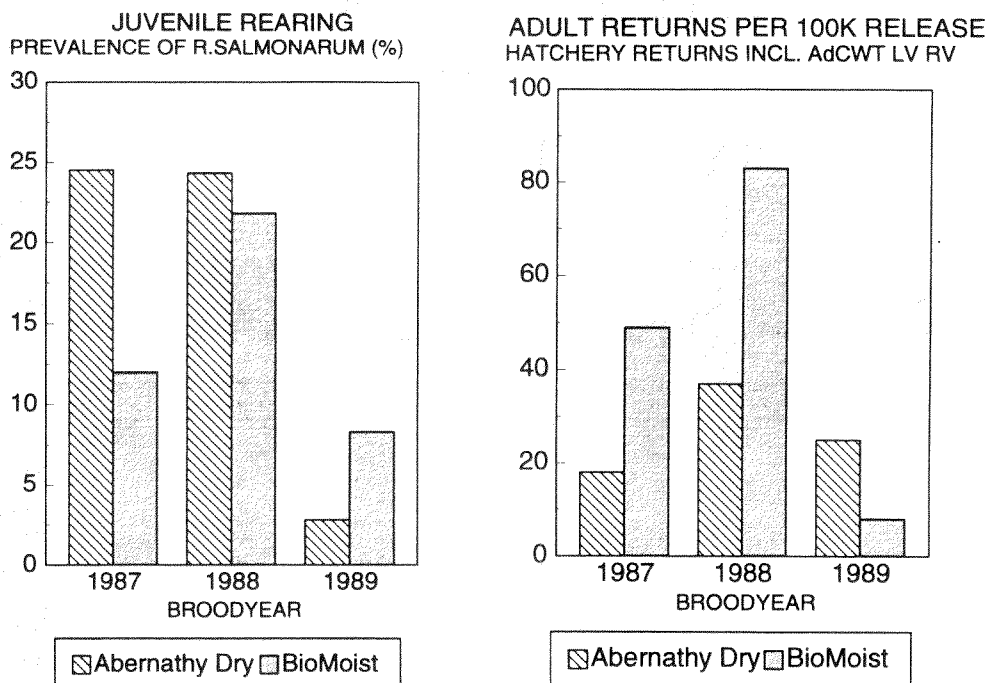


Figure 9. Diet, bacterial kidney disease, and adult yield.

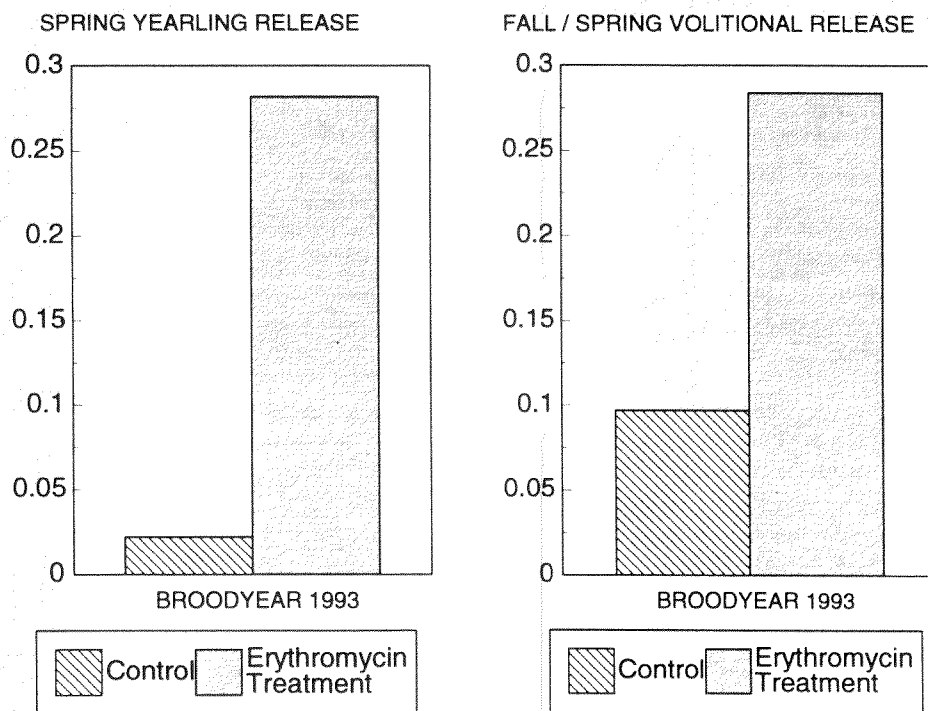


Figure 10. Hatchery adult recoveries (%) for medicated feed study.

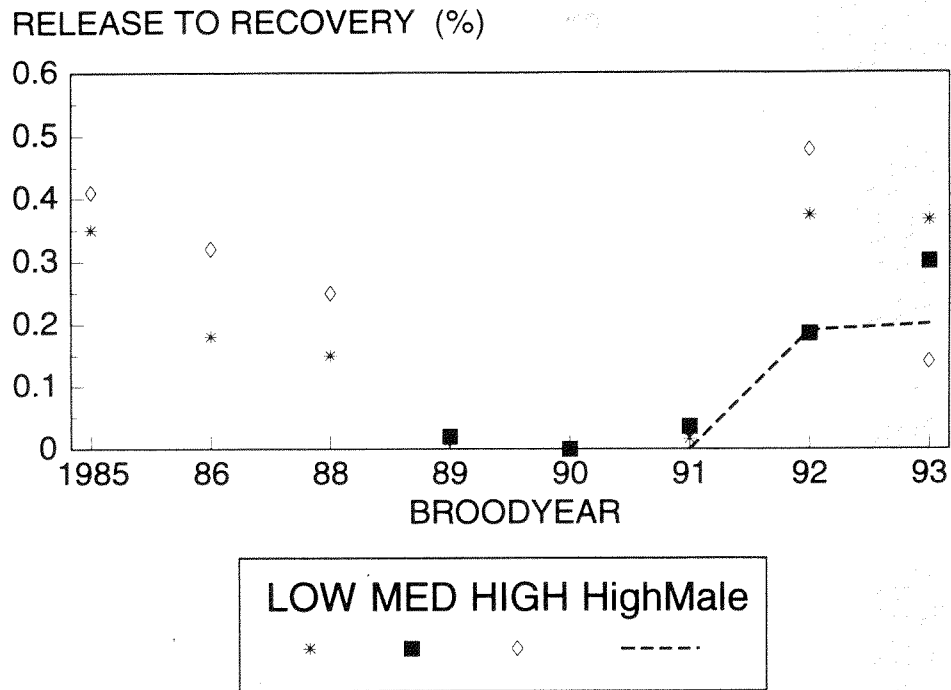


Figure 11. ELISA segregation of spawners with O.D. < 0.5 and survival of progeny to adult.



# PRACTICAL FISH CULTURE AT GROVERS CREEK SALMON HATCHERY

Paul Dorn & Randi Thurston

*Suquamish Tribal Fisheries Department, PO Box 498, Suquamish, WA 98392*

Andy Appleby

*Washington Department of Fish and Wildlife, 600 Capitol Way N, Olympia, WA 98501-1091*

Jay DeLong & Sharon Lutz

*Northwest Indian Fisheries Commission, 6730 Martin Way E, Olympia, WA 98516*

## Introduction

Fall chinook have been hatchery reared since 1895 to mitigate for declining local natural abundance and/or supplement or create fisheries in Washington State (WDFW). Hatchery survival and fishery contribution rates have ranged from low to high, compared to wild stocks, as we have learned the intricacies of nutrition, fish health, stock genetics, and the natural and artificial environments' influence on fish behavior (WDFW, hatchery records). Hatchery cultural practices continue to evolve as we incorporate new knowledge into our programs. This paper reviews the Suquamish Indian Tribe's (SIT) fall chinook salmon enhancement program at Grovers Creek Hatchery, begun in 1978 with the cooperation with the Washington Department of Fish and Wildlife (WDFW). The program was designed to restore Tribal chinook fisheries on the west side of central Puget Sound, adjacent to the Kitsap Peninsula. There are no native runs of fall chinook in this area.

Grovers Creek Hatchery received eyed eggs from WDFW of Soos Creek origin between 1978 to 1981. Adults returning to the hatchery in 1982 represented the first mixed age class used to supply 100% of the hatchery broodstock. Eggs surplus to SIT needs are delivered to WDFW for in-State programs, or sold when the in-State production goals are attained. The hatchery annually releases an average of 537,000 smolts that are in proportion to the return timing. Off-station rearing ponds were established, beginning in 1982, that are supported by the hatchery broodstock. The chinook smolts released from the three off-station rearing sites are represented in proportion to the adult run timing spectrum. WDFW provides the balance of chinook fry to meet off-station production goals in years Grovers Creek Hatchery broodstock returns are low. The SIT fishery excludes targeting the hatchery broodstock, focusing the Tribal

terminal fishery exclusively on the off-station rearing ponds. Grovers Creek Hatchery fall chinook have been coded wire tagged continuously since brood year 1981.

## Hatchery Management

Adult fall chinook salmon return to Grovers Creek Hatchery in mid-September and continue until the end of October each year. The adults return to the same earthen pond in which they were reared. The peak of the return is the last week of September and first week of October. Adults are seined and spawned weekdays on Monday, Wednesday, and Friday. Grovers Creek Hatchery's water supply limits the number of fall chinook eggs to two million. The SIT program goal is 3.2 million eggs and is satisfied by WDFW incubating one million eggs at Minter Creek Hatchery and the Mid Sound Fisheries Enhancement Group incubating a quarter million eggs at Burley Creek Hatchery.

Grovers Creek adult fall chinook are spawned throughout the entire run. Typically, jacks (2-year-old males) are separated from the adults used for broodstock and contribute to less than 5% of the spawning population. Grovers Creek broodstock is obtained by stripping eggs from individual females into a small bucket, with sperm from two different males added. The second male is used to increase the probability that the sperm is viable. Stream water is introduced to the bucket and the rinsed eggs are transferred to a 5 gallon bucket for water hardening in a 100 ppm iodophor solution for one hour.

Chinook eggs that are transferred off-station are delay-fertilized with five males stripped into individual ziplock bags and thirty females stripped into individual bags inside 30 gallon buckets. All delayed gametes are transferred on ice. The Grovers Creek water hardened eggs are transferred to heath trays or deep matrix boxes for incubation. Pathogen and silt-free 10° C groundwater

is used to incubate all Grovers Creek Hatchery fall chinook eggs. A 1:600 formalin treatment is applied three times a week via a 15 minute pumped treatment. The fungus treatment is discontinued at 425 temperature units, after shocking and egg picking but before the eggs hatch. Swim-up fry are ponded indoors into circular ponds for initial feeding, then transferred outdoors into two ponds for rearing with ambient temperature Grovers Creek water. The chinook fry are introduced into the ponds throughout January. The pond temperature averages 2° C during January, slowing the growth rate of the early spawned chinook fry so they are not significantly larger than the chinook fry spawned late in the run. Grovers Creek provides an average of 2500 gpm in January - February, but stream flow through the rearing pond diminishes to 300 gpm by late May.

Grovers Creek Hatchery chinook fry are reared in two random groups, with half ponded into a 9,100 ft<sup>3</sup> cement pond and the other half ponded into a 29,000 ft<sup>3</sup> unlined earthen pond. Approximately 200,000 of the 9,100 ft<sup>3</sup> group are coded wire tagged (CWT) at 2.2 gms. The CWT fry are released into the earthen pond after tagging and complete their rearing with their untagged cohort. A moist diet is fed at the manufacturers suggested rate, with changes in pellet size dictated by the smallest fry in the population. No grading or handling of the fry, except for weight samples and fish health inspections, occurs for the duration of their freshwater rearing. Outlet screens are removed from the pond when the chinook reach 5 gms to allow volitional outmigration, typically in late April. Feeding continues until late May with the use of a smolt counter, borrowed from the U.S. Fish and Wildlife Service, positioned in the fish ladder to enumerate outmigration progress. The station target is to produce a 9 gm smolt. The rearing pond is 100 meters from saltwater at high tide.

Avian predation is controlled by the use of a 5 cm knotless polypropylene net stretched over the entire pond. The net is suspended over three cables running the length of the pond. The center cable is the highest and can be raised or lowered by a manual boat winch to prevent snowload damage. An electric fence set 8 cm above the ground eliminates river otter predation on the young growing chinook fry.

#### Fish Health

The health status of Grovers Creek chinook has been monitored since 1981 (Table 1). Adult broodstock are

screened for the presence of viral or bacterial pathogens and juvenile fish are monitored on a monthly basis to assess general fish health and identify any potential problems occurring in the population.

To date, adult broodstock have been relatively disease free. Inspection examinations of returning adults have identified low levels of the bacterium *Renibacterium salmoninarum*, the causative agent of bacterial kidney disease. This pathogen has also been identified in juveniles at very low levels but no mortality has been attributed to this disease. No viral pathogens have been isolated from this stock of fish, however, between 1991 and 1996, two non-pathogenic viral agents (*reovirus*, *paramyxovirus*) were isolated during normal adult inspection screening.

Juvenile chinook reared at Grovers have had a variety of parasites, bacteria and environmental conditions associated with them. Water flow constraints and variable environmental conditions have made fish rearing challenging. Between 1981 and 1990, environmental gill disease and bacterial gill disease were major problems that resulted in significant losses. These conditions were brought on by a variety of conditions including decreased water flows, increased water temperatures, low dissolved oxygen levels and overcrowding. The parasites, *Ichthyobodo* and *Ichthiophthirius multifiliis* have also caused some elevated mortality over the years. All these diseases were managed through the application of chemical treatments.

In an effort to improve the rearing environment, some rearing strategy changes were implemented between 1989 and 1991. These included providing aeration to the rearing pond, adding an additional well water source to supplement stream flow and establishing an in-house fish health monitoring program. In 1991, the Grovers facility obtained a microscope and personnel were trained to evaluate gill condition and identify gill disease bacteria. This allowed for frequent gill condition monitoring. These three factors combined have made an extreme difference to this program. Environmental and bacterial gill disease are no longer the problem they once were. Of course, these conditions can still be observed but they are kept in balance without experiencing the high mortalities that had occurred in the past. Currently, chemical treatments are used infrequently and due to the increased monitoring capabilities most potential problems are detected early and managed accordingly.

Practical Fish Culture at Grovers Creek Salmon Hatchery

Table 1. Grovers Creek Facility - Fall Chinook Fish Health Observations

(includes any detection of a pathogen or condition, but not necessarily that a disease condition was associated)

Monitoring Year	Juveniles	Average Mortality	Treatment	Adult Inspections
1981				<i>Renibacterium salmoninarum</i>
1982	Bacterial gill disease	Moderate (0.031-0.10%/day)	Hyamine.	
1983	<i>Ichthyobodo</i> (Costia)	Normal to low (<0.01 - 0.03%/day)		<i>Renibacterium salmoninarum</i>
1984	Environmental gill disease <i>Ichthyobodo</i> (Costia), <i>Epistylis</i>	Normal to low (<0.01 - 0.03%/day)		<i>Renibacterium salmoninarum</i>
1985	Environmental gill disease Bacterial gill disease, <i>Ichthyobodo</i> (Costia)	Low to moderate (0.011-0.10%/day)	Diquat and formalin for gill disease and costia.	<i>Renibacterium salmoninarum</i>
1986	Environmental gill disease, <i>Phoma</i> sp.	Normal to low (<0.01 - 0.03%/day)		<i>Renibacterium salmoninarum</i>
1987	<i>Ichthyobodo</i> (Costia), <i>Ichthyophthirius multifiliis</i>	Low to moderate (0.011-0.10%/day)	Formalin for costia and ich.	
1988	Environmental gill disease Bacterial gill disease, <i>Ichthyobodo</i> (Costia)	Moderate to high (0.31%>0.11%/day)	Diquat for gill disease.	<i>Renibacterium salmoninarum</i>
1989	Environmental gill disease Bacterial gill disease <i>Aeromonas</i> sp., <i>R. salmoninarum</i>	Moderate to high (0.31%>0.11%/day)	No chemical treatments applied.	<i>Renibacterium salmoninarum</i>
1990	Environmental gill disease Bacterial gill disease, <i>Epistylis</i> , <i>Ambiphrya</i>	Low to moderate (0.011-0.10%/day)	Diquat for gill disease.	
1991	Coagulated yolk syndrome Environmental gill disease Bacterial gill disease <i>Ichthyobodo</i> (Costia)	Normal to low (<0.01 - 0.03%/day)	Prophylactic diquat treatment initiated prior to tagging for gill disease.	Reovirus
1992	Environmental gill disease <i>Ichthyobodo</i> (Costia), <i>Hexamita</i> sp. Coagulated yolk syndrome Bacterial kidney disease	Normal (<0.01%/day)	Prophylactically treated for gill disease with KMnO4. Formalin used to treat costia.	
1993	Environmental gill disease Bacterial gill disease <i>Pseudomonas</i> sp., <i>Epistylis</i> , <i>Ambiphrya</i> Coagulated yolk syndrome	Normal (<0.01%/day)	No chemical treatments applied	Paramyxovirus
1994	Coagulated yolk syndrome	Normal (<0.01%/day)		
1995	<i>Flavobacterium psychrophilum</i>	Normal (<0.01%/day)	No chemical treatments applied	Reovirus
1996	Environmental gill disease Bacterial gill disease <i>Flavobacterium psychrophilum</i> <i>Ambiphrya</i> , <i>Phoma</i> sp. Coagulated yolk syndrome	Normal to low (<0.01 - 0.03%/day)	No chemical treatments applied	Paramyxovirus



### Production and Adult Returns

Grovers Creek Hatchery fall chinook have been raised in a low capital cost facility within a suburbanizing watershed. The earthen pond approach produced quality smolts for the first seven years, but declining water quality and quantity (seasonally) impacted production (Figure 1). Environmental and bacterial gill disease decreased production in 1987, 1988, and significantly in 1989. Aggressive aeration (with a 5 hp blower and airstone matrix suspended just off the pond bottom), well water supplementation during low stream flows, and application of a soil bacteria solution at water temperatures above 10° C restored fish health and smolt quality at release. Station production was low in 1994 due to low returns of adults to Grovers Creek Hatchery.

The average Grovers Creek Hatchery rack return is 2,500 adults per year, but has varied significantly (Figure 2). Scales are removed from 200 adult fall chinook each week at the hatchery rack for age analysis. The results are used to forecast future runs, both to the hatchery and the off-station rearing ponds and to evaluate changes to the hatchery population over time. 100% of Grovers Creek fall chinook are inspected for adipose clips and heads are processed in the spawning shed. The length and weight of each fish is taken and recorded on both the scale card and hatchery field logs.

### Coded Wire Tag Study Results

Expanded coded wire tag recovery data were obtained from the Pacific States Marine Fisheries Commission (PSMFC) and summarized by the NWIFC Coded wire tag Recovery and Analysis System (CRAS). Percent recoveries of brood years 1981 to 1991 were summarized by area and fishery, and survival rate estimates were calculated (Table 2). Estimates of total marine catch, and total catch by area and fishery, were generated (Table 3).

### Straying

Recoveries of coded-wire tags allow a glimpse at of Grovers Creek fall chinook straying, as well as straying to Grovers Creek Hatchery. Figure g shows all Grovers Creek fall chinook freshwater recovery locations from 1982 to 1985. Figure h shows the hatchery origins of BY 1982-1995 fall chinook recovered at Grovers Creek Hatchery.

### Observed Trends in Hatchery Broodstock Size

Since the body size of adults returning to the hatchery can provide an integrated assessment of the environmental and genetic factors that have affected the fish (Gall 1987), a study was conducted to determine the trend for body weight and length of Grovers Creek Hatchery fall chinook returning to the hatchery between 1986-96.

For each sex, 3 and 4 year old fish were analyzed separately. A systematic random sample size of 50 was determined necessary to estimate mean weight and length. This sample size was not available for both sexes of each year class because of low returns in 1991, 1992, and 1994 and inadequate data in 1990.

The null hypothesis that the observed mean weight and length from a random sample of the population would not significantly change between 1986-96 was tested against the alternative hypothesis that the observed mean weight and length from a random sample of the population would significantly decrease between 1986-96. The null hypothesis was rejected for 6 of the 8 trends analyzed (all but weight of 3 year old males and length of 4 year old males) (Figures x and y). Therefore, we concluded that fish lengths and weights of the other 6 groups decreased over the time period of the study, but it could not be demonstrated by this study that weight of 3 year old males and length of 4 year old males decreased over time.

The trend toward decreasing body size of Grovers Creek Hatchery fall Chinook corresponds to that seen in other studies of North Pacific salmon (Bigler et al. 1993, Healey 1986, Ricker 1995). Possible causes for the decrease in body size of the Grovers Creek fall chinook stock include ocean climate conditions, density dependent competition, and genetic changes due to size selective fishing or hatchery management practices.

Beamish (1993) found that an increase in the intensity of the Aleutian low pressure system correlated well with strong year classes and above average survival of salmon. But, an inverse relationship between population abundance and mean body size occurred during the same period. This suggests that there may a limit to the salmon sustaining resources of the ocean (Bigler et al. 1996).

Pacific salmon enhancement programs have assisted in the near doubling of salmon harvests over the past two decades in the North Pacific. During the period of favorable ocean climate conditions from 1973-1993, 45 of 47 North Pacific salmon populations studied by Bigler

et al (1996) decreased in average body size. Washington chinook salmon stocks caught in the troll and Columbia River fishery declined in average body size 10.09% - 46.70% between 1976-93, possibly due in part to increasing hatchery releases causing a reduction in the available food supply through density dependent competition.

Ricker (1995) concluded the mean weight of chinook salmon caught by commercial trolling in Puget Sound between 1975-80 decreased ~1.5 kg, and showed little recovery to 1990, although chinook caught between 1985-1987 were larger. He suggests that since early maturing fish grow faster than those that mature at an older age, the selection of larger, slower growing older fish by a fishery may affect the heritable aspects of the growth rate and age at maturity causing a population to shift toward faster growth and younger age at maturity.

Despite using strict genetic conservation measures, hatcheries risk genetic change because their populations are relatively small and closed (Gall 1987), mainly due to genetic drift, the random loss of certain genes in small populations and to inbreeding (breeding closely related individuals). Generally, the greater the inbreeding, the more pronounced the reduction in viability, growth, survival and fecundity (Tave 1986 and Gall 1987).

In contrast to our observations of Grovers Creek fall chinook, a study of fall chinook salmon produced at four Washington State Department of Fish and Wildlife hatcheries found no decrease in length over brood years 1971-1992 (Vander Haegen and Appleby 1996). Unlike the Grovers Creek study, however, mean lengths in their study were calculated for males and females combined.

A second analysis was performed using mean lengths of Grovers Creek fish with sexes combined for brood years 1985-1991. The results are shown in Figure 7, along with WDFW Soos Creek Hatchery study results. WDFW data were not available for brood year 1989. Grovers Creek results were similar to those of the WDFW study for this interval for this combined-sex study. The data suggest a decrease in length, but the trend line plotted is not statistically significant. It is not known if analysis of the WDFW fish by sex would change their findings.

The observed decreases in size of 6 of the 8 Grovers Creek Hatchery fall chinook age-sex combinations may not reflect the long-term trend because only 11 years of data were analyzed.

### Off-station Rearing Pond Production and Fishery Contribution

Grovers Creek Hatchery production supports three off-station rearing ponds that contribute to an important Suquamish terminal fishery (Figure 3). All three sites are operated primarily with sports club volunteers and have limited operations and maintenance funding. WDFW provides fry to make up Grovers Creek shortfalls as well as providing most of the fish food. No hatchery personnel live on-station and emergencies are handled by volunteer phone tree basis. The risks of this approach were demonstrated by a massive fish kill in 1992 due to a clogged intake screen.

### Literature Cited

- Beamish, R.J. 1993. Climate and exceptional fish production off the west coast of North America. *Can. J. Fish. Aquat. Sci.* 50: 2270-2291.
- Bigler, B.S., D.W. Welch, and J.H. Helle 1996. A review of size trends among North Pacific salmon (*Oncorhynchus* spp.). *Can. J. Fish. Aquat. Sci.* 53: 455-465.
- Gall, G.A.E. 1987. Inbreeding. Pp. 47-87. In N. Ryman and F. Utter, eds. *Population Genetics and Fishery Management*. University of Washington Press, Seattle, WA.
- Healey, M.C. 1986. Optimum size and age at maturity in Pacific Salmon; effects of size selective fisheries. *Can. Spec. Publ. Fish. Aquat. Sci.* 89.
- Ricker, W.E., 1995. Trends in the average size of Pacific salmon in Canadian catches, p. 593-602. In R.J. Beamish ed. *Climate change and northern fish populations*. *Can. Spec. Publ. Fish. Aquat. Sci.* 121.
- Tave, D. 1986. *Genetics for fish hatchery managers*. AVI Publishing Company, Inc. Westport, Connecticut.
- Vander Haegen, G. and A. Appleby. 1996. Size trends in coho and fall chinook salmon produced at WDFW hatcheries.

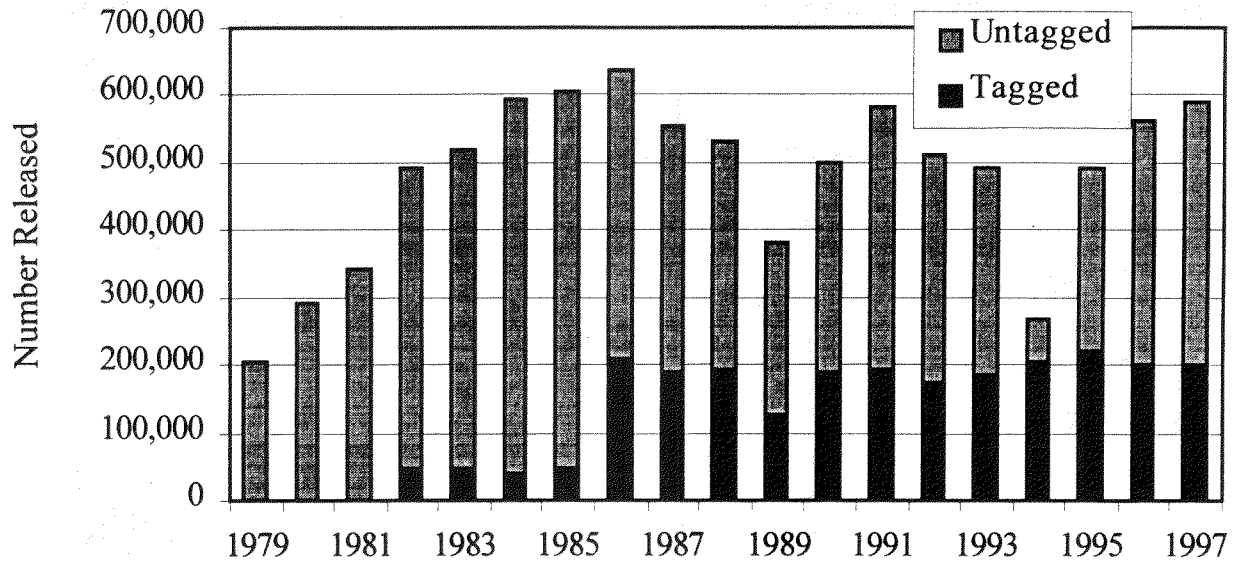


Figure 1. Grovers Creek Hatchery fall chinook releases with coded wire tagged component.

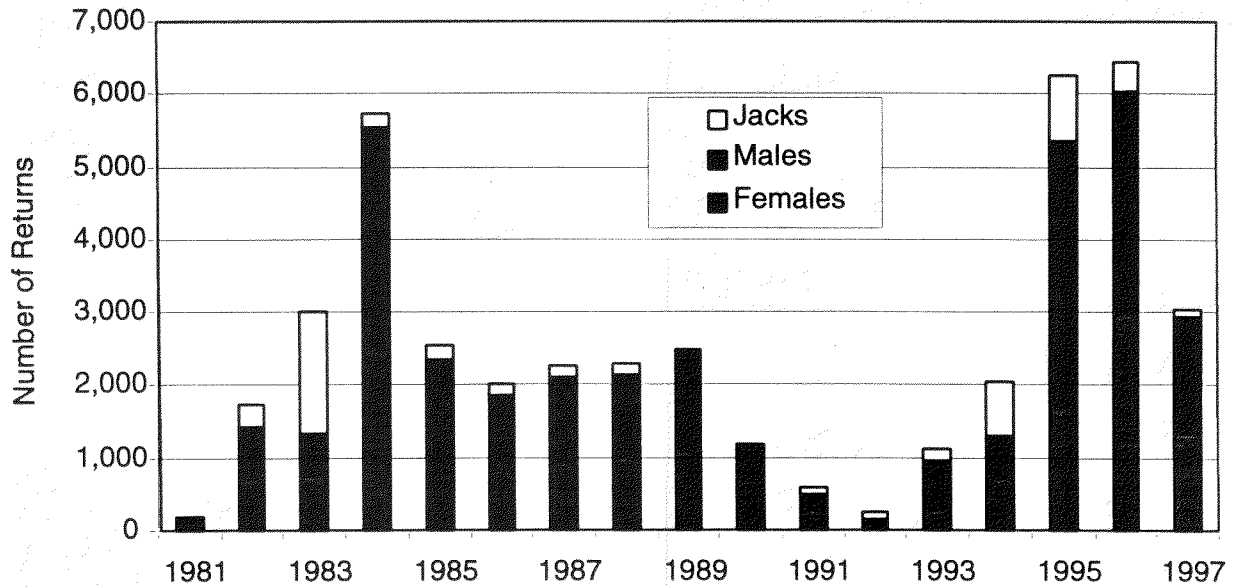


Figure 2. Grovers Creek Hatchery adult fall chinook return by sex.

Table 2. Estimated Grovers Creek fall chinook survival rates and CWT recoveries by area and fishery

Brood Year	# Tagged	# Released	# Est. Recoveries	Est. % Surv.	Alaska	Canada	Juan de Fuca Sport	Juan de Fuca Comm.	WA Coast & South	N Puget Sound Sport	N Puget Sound Comm.	Mid Puget Sound Sport	Mid Puget Sound Comm.	S Puget Sound Sport	S Puget Sound Comm.	Hatchery
1981	47,471	489,965	1,524.5	3.2	0.2	17.3	3.4	1.7	0.3	0.6	1.0	25.9	7.1	3.9	0.6	37.6
1982	45,284	520,800	345.8	0.8	0.0	32.0	2.9	1.3	2.0	0.7	1.1	18.9	1.3	2.8	0.6	29.9
1983	40,324	594,000	307.9	0.8	0.0	21.5	1.9	11.2	0.7	0.0	0.0	13.2	11.8	0.0	0.0	39.0
1984	45,907	606,500	602.8	1.3	0.3	31.0	4.2	9.9	1.3	0.0	0.2	11.3	9.8	0.8	0.2	31.2
1985	207,155	637,032	1,367.2	0.7	0.2	20.7	5.8	11.5	2.5	0.0	2.8	2.5	4.9	0.0	0.4	48.0
1986	187,757	554,163	3,45.2	1.6	0.0	18.1	5.6	13.3	1.7	0.7	0.5	10.7	9.7	0.7	0.5	38.3
1987	193,906	531,351	911.2	0.5	0.0	22.2	6.4	16.3	0.7	1.0	0.4	7.4	7.1	0.3	0.3	36.2
1988	124,626	380,239	130.2	0.1	0.0	17.7	6.1	17.0	0.0	3.2	0.0	8.2	5.1	0.0	1.5	41.2
1989	187,640	501,391	303.6	0.2	1.5	16.1	16.3	12.4	2.0	0.0	0.0	6.4	6.7	0.0	0.6	32.6
1990	193,496	580,288	1,435.7	0.7	0.1	22.0	5.0	6.3	0.3	2.4	0.2	10.6	4.3	0.1	0.0	48.6
1991	174,949	509,815	477.1	0.3	0.5	14.0	4.1	0.9	0.0	1.8	0.0	12.4	4.8	0.8	0.0	55.8
Average					0.3	21.2	5.6	9.3	1.0	0.9	0.6	11.6	6.6	0.8	0.4	39.8

Table 3. Estimated total catch of Grovers Creek fall chinook by area, including escapement.

Brood Year	Total Catch	Alaska	Canada	Juan de Fuca Sport	Juan de Fuca Comm.	WA Coast & South	N Puget Sound Sport	N Puget Sound Comm.	Mid Puget Sound Sport	Mid Puget Sound Comm.	S Puget Sound Sport	S Puget Sound Comm.	Escapement
1981	15,681	31	2,726	539	275	44	102	155	4,075	1,118	608	99	5,909
1982	3,719	0	1,274	116	51	79	29	44	750	53	110	23	1,190
1983	4,499	0	944	84	507	31	0	0	600	533	0	0	1,800
1984	7,964	21	2,472	334	789	100	0	13	900	777	62	15	2,481
1985	4,155	7	872	243	484	87	0	119	105	204	0	16	2,018
1986	8,901	0	1,629	501	1,193	91	63	43	962	869	66	43	3,441
1987	2,448	0	555	159	407	12	25	10	186	177	7	6	904
1988	397	0	70	24	67	0	13	0	33	20	0	6	164
1989	758	12	130	132	100	8	0	0	52	54	0	5	265
1990	4,295	6	949	213	273	9	102	10	455	184	3	0	2,091

Region	Recovery Location	# Estimated CWT Recoveries
Puget Sound	Baker River	1
	Coulter Creek Hatchery	1
	Hupp Springs Rearing Facility	1
	Issaquah Creek	1
	Minter Creek	1
	Tulalip Salmon Hatchery	1
	Issaquah Hatchery	2
	Newaukum Creek (Green R)	3
	Burley Creek	6
	Capitol Lake Rearing Facility	6
	Soos Creek Hatchery	4
	Minter Hatchery	5
	McAllister Hatchery	7
	Garrison Hatchery	16
	Grovers Creek Hatchery	6478 (99.2% of total)
TOTAL		6533

Table 4. Freshwater recovery locations of Grovers Creek fall chinook, 1985-1995.

Region	Recovery Location	# Estimated CWT Recoveries
Canada	Chemainus River	2
	Cowichan River	1
Columbia River	Little White Salmon	1
	Cowlitz Hatchery	1
Hood Canal	Fox Island	1
	Sund Rock Hatchery	1
	Big Beef Hatchery	2
	Quilcene Hatchery	3
Strait of Juan de Fuca	Elwha	1
	Garrison Hatchery	2
Puget Sound	Allison Springs	1
	Portage Bay Hatchery	1
	Grovers Creek Hatchery	6478 (99.7% of total)
TOTAL		6495

Table 5. Origins of fall chinook recovered at Grovers Creek Hatchery

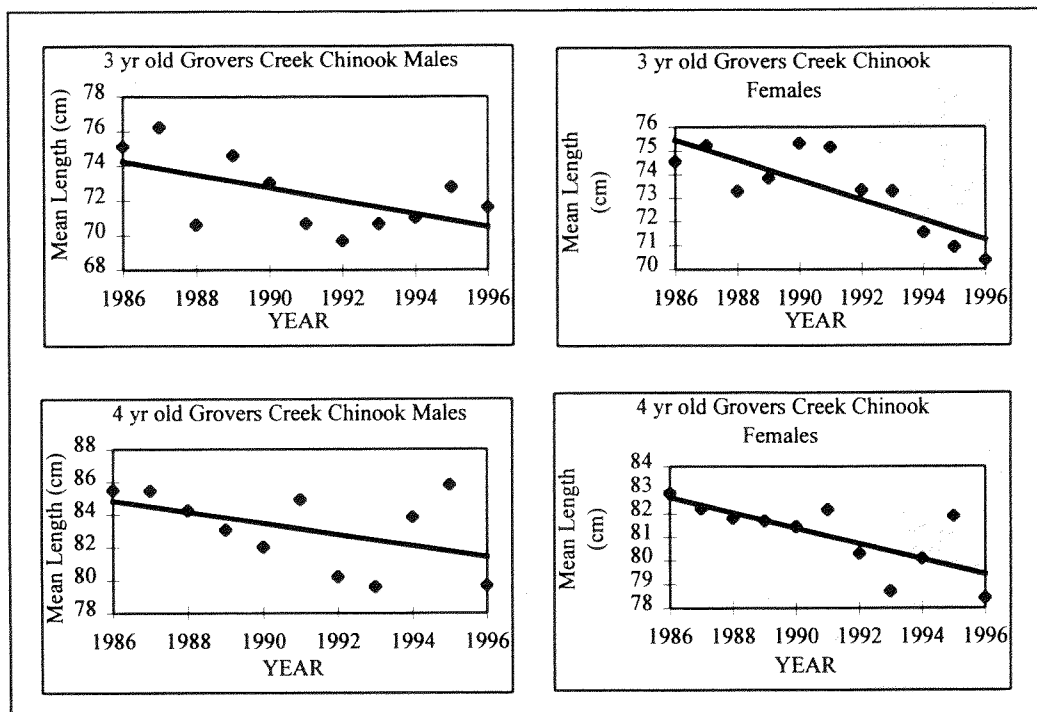


Figure 3. Changes in length of returning Grovers Creek fall chinook, by age and sex, for years 1986-1996. The null hypothesis of no difference was rejected for all but the 4-year-old males.

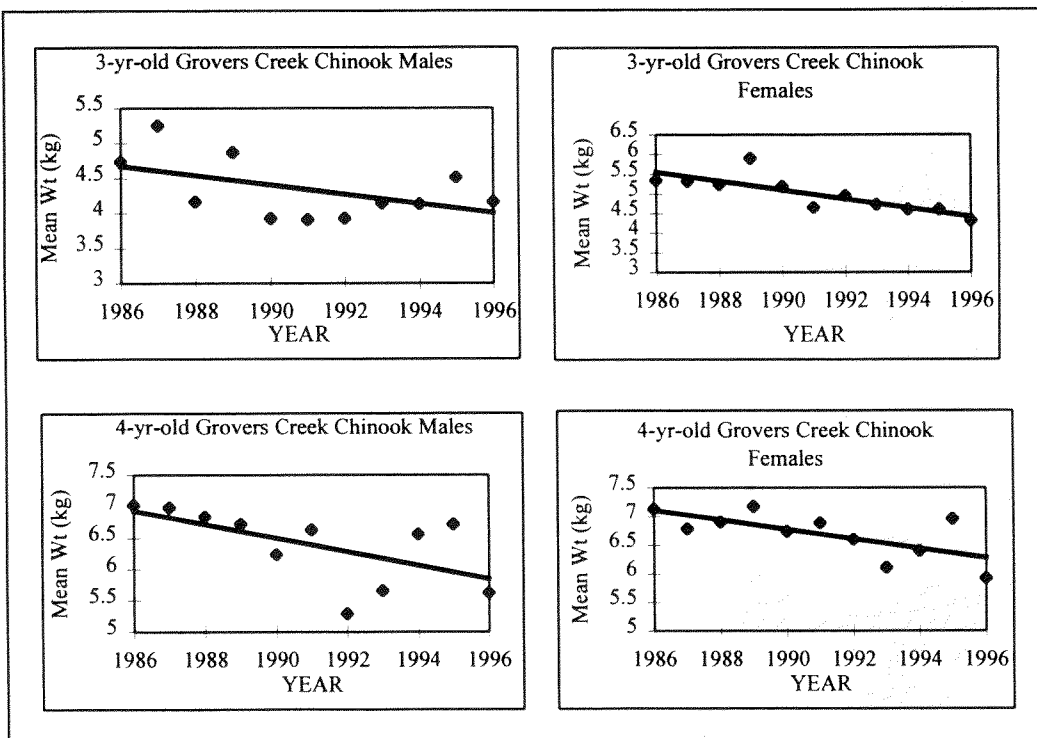


Figure 4. Changes in weight of returning Grovers Creek fall chinook, by age and sex, for years 1986-1996. The null hypothesis of no difference was rejected for all but the 3-year-old males.

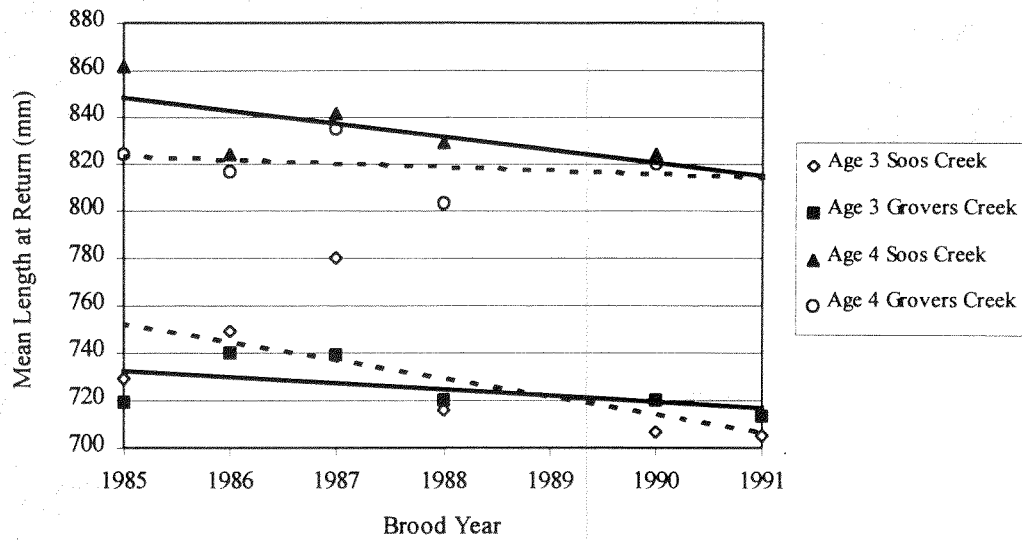


Figure 5. Changes in Mean Length of Grovers Creek and Soos Creek Fall Chinook, Brood Years 1985-1991.

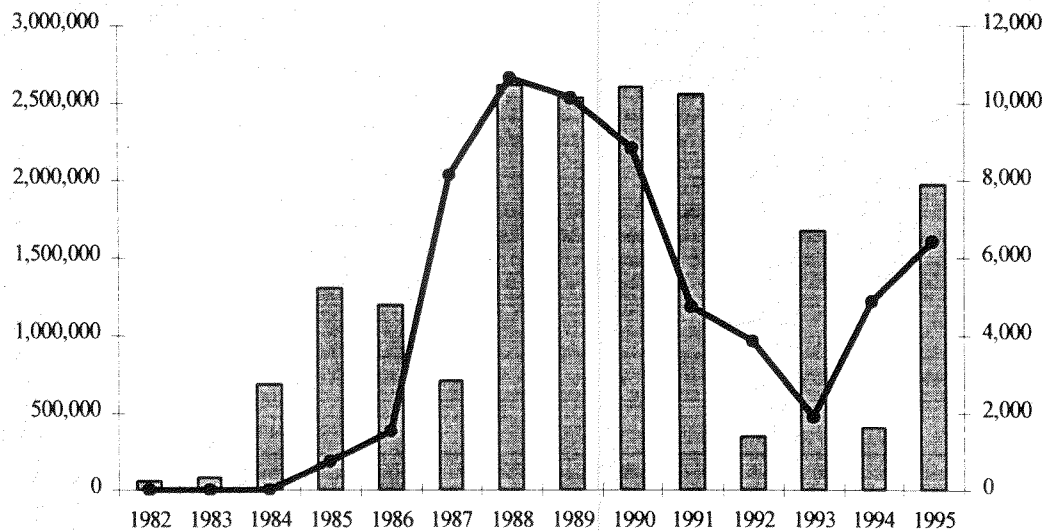


Figure 6. Number of Suquamish Off-station Fall Chinook Smolts Released (bars) and Number of Chinook Caught in Tribal Terminal Fishery (line).

Practical Fish Culture at Grovers Creek Salmon Hatchery

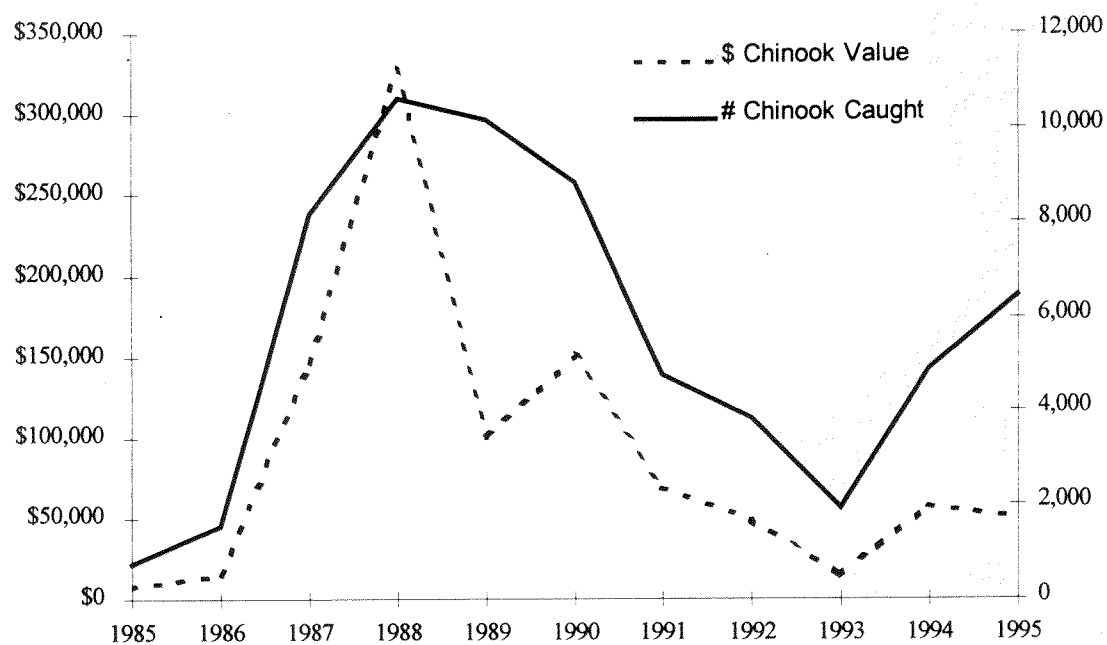


Figure 7. Suquamish Tribe Area 10E commercial chinook harvest and value, 1985-1995.





## **Aquaculture in Far East Russia: Kamchatka Peninsula and Sakhalin Island**

Jack Hurst <sup>1</sup>

*Oregon Department of Fish and Wildlife*

Some of the Kamchatka and Sakhalin regions major natural resources and industries are oil, timber, and fishing, and are perceived to be the economic base for the future. Although oil resources are currently being developed, logging and fishing industries have been active for many years. Many other industries that were counted on for support within the regions, such as paper products and the military, were abandoned around 1991. This economic collapse, as I observed, has caused many hardships to overcome for these people. Most jobs are non-existent or hard to come by in most areas, except the larger cities. The abundance of salmon in local streams for these people is then used for subsistence and, by permit, some economic gain.

Aquaculture, related facilities and programs, in the far East of Russia, have been in existence for many years.

These facilities are government operated by regional fish protection agencies called "Ribvod". These agencies are structured somewhat like our State and National Fish & Wildlife agencies. These "Ribvod's" are empowered to protect and enhance their regional fish and marine mammal resources. Some stations are over 80 years old. Most facilities have gone, or are now going, through reconstruction to meet the demands put on the resource. Most demands stem from ocean and instream commercial fishing and subsistence needs.

This presentation focuses on these fish culture facilities, and the complexity of fish protection and the dedication of those who are empowered to protect and enhance this resource in these regions.

---

<sup>1</sup> Jack elected to only have the abstract of his talk included in the Proceedings.

Hurst



# Posters

## **Propagating Juvenile Fall Chinook in Michigan Raceways at Umatilla Hatchery**

Wes Stonecypher, Shannon Focher, Warren Groberg, Jr., Mike Hayes, Hack Hurst, Sam  
Onjukka, Karen Waln, and Randall Winters

*Oregon Department of Fish and Wildlife*

In addition to the formal presentations, Shannon Focher presented a well executed poster on propagating fall chinook at Umatilla Hatchery using the raceway technology developed in the state of Michigan.



## COMMERCIAL EXHIBITORS

COMPANY	REPRESENTATIVE	PHONE	FAX	E-MAIL ADDRESS
HARPER BRUSH CO.	KEN TAYLOR	425-255-2074	425-235-6709	
THE MALLORY CO.	STEVE GAUTHIER	360-636-5750	360-577-4244	info@malloryco.com
COMMON SENSING	BRIAN D'Aoust	208-266-1541	208-266-1428	comsen@dmf.net
MAGIC VALLEY HELI-ARC	LINDA OWENS	208-733-0503	208-733-0544	
CHRISTENSEN NET WORKS INC.	SCOTT CHRISTENSEN	360-384-1446	360-384-1446	
JENSORTER	GREG JENSEN	541-389-3591	541-389-0050	jensorter@transport.com
INTERMOUNTAIN WEIGHING SYSTEM, INC.	TOM BRADLEY/ CHRISTINE BRADLEY	208-362-3667	208-362-5285	tomlbs@aol.com
WESTERN CHEMICAL INC.	RON SECOR	800-283-5292	360-384-0270	wci@premier1.net
NELSON & SONS, INC./ SILVER CUP FISH FEED	CHRIS NELSON/ JERRY ZINN	800-521-9092	801-266-7126	silvercup@xmission.com
NORTHWEST SCALE SYSTEMS INC.	PHIL CHAPPLE/ TERRY HOFFMAN	206-575-0074	206-575-0082	chappie27@aol.com
PRA MANUFACTURING LTD	WAYNE GORRIE	250-754-4844	250-754-9848	pramfg@island.net
J.L. EAGAR, INC.	STAN BELL	801-292-9017	801-295-7569	
MARISOURCE/ FLEX-A-LITE CONSOLIDATED, INC.	EDDY WILLINGHAM	253-922-2700	253-922-0226	flex@flex-a-lite.com
MOORE-CLARK	RON MALNOR/ STEVE BIGGIO	604-325-0302 425-744-4500	604-325-2884 425-744-6619	
RANGEN, INC.	JERRI FULLERTON	208-534-6421	208-543-4698	ragenff@magiclink.com
BIOPRODUCTS	RUSS FARMER/ BRUCE BUCKMASTER	503-861-2256	503-861-3701	
POINT FOUR SYSTEMS INC.	TJARDA BARRATT	604-936-9936	604-936-9937	tjardab@pointfour.com
WARREN WATER BROOM	DELL WARREN	503-458-6694		
CASCADE MACHINERY & ELECTRIC	GARY CARTER	503-650-2317	503-650-2660	
VMG INDUSTRIES INC.	BRUCE MARSHALL	970-242-8623	970-243-3563	
INNOVATIVE COATING SOLUTIONS, INC.	JAY GLOVER	360-695-7199	360-883-3566	ics@worldaccessnet.com
EMA- ENGINEERED PRODUCTS DIVISION	T.R. GREGG	541-929-3225	541-929-2279	greggt@peak.org
LFS, INC.	TERRY CRUMP	360-734-3336	360-734-4058	bham@lfsinc.com
FAMILIAN NORTHWEST	VICTOR CLEMENTS	360-835-2129		





## Door Prize List

<u>Prize</u>	<u>Recipient</u>	<u>Donor</u>
Fish Print	Susan Gutenberger- USFWS	Western Chemical
2 Bottles of Wine and a Cap	Judy Urrutia- CDEG	Lambs Thriftway/Marisource
Coleman Lantern	John Schmitz- ODFW	Common Sensing
Jansport Day Pack	Steve Olhausen- USFWS	Western Chemical
Cargo Net	Paul Kaiser- USFWS	Christensen Net Works
Fish Print	Todd Doughton- ODFW	Western Chemical
2 Bottles of Wine and a Cap	Denise McCarver- WDFW	Lambs Thriftway/Marisource
Bushnell Binoculars	Joe Hulburt- ODFW	Western Chemical
Leatherman Micro Tool	Rodney Roeder- USFWS	Common Sensing
Katheryn Kostow Print	Paul Dorn- Suquamish Tribal Fisheries Department	Kathryn Kostow
Jacket	Donovan Ward- CDEG	The Mallory Company
Gerber Knife	Roger Warren- ODFW	Bioproducts
Mouse Pad and Sweatshirt	Mark Bushman- ODFW	Leavenworth Hatchery
2 Bottles of Wine and a Cap	John Holmes- USFWS	Lambs Thriftway/Rangens
Silver Cup Sweatshirt	John Halver- University of Washington	Nelson & Sons/ Silver Cup Feeds
Mag Light Flashlight	Charmane Ashbrook- WDFW	Common Sensing
Jansport Waistpack	Mike Stratton- ODFW	Western Chemical
2 Bottles of Wine, CD, and Cap	Karl Schearer- NMFS	Lambs Thriftway/Utah Dept. Fish & Game/ Marisource

<u>Prize</u>	<u>Recipient</u>	<u>Donor</u>
T-shirt and Cook Book	Dale Diog- ODFW	Sunshine Coast Salmon Enhancement Society/ Martha Klontz
Leatherman Micro Tool	Andy Van Scoyk- Rowdy Creek Fish Hatchery	Common Sensing
Jansport Day Pack	Doug Dysart- USFWS	Western Chemical
Ghirardelli Old Fashion Chocolate Truck	Andrea Pinkerton- WDFW	Argent Chemicals
Coleman Lantern	Jim Barfoot- Trout Lodge	Common Sensing
2 Bottles of Wine and a Cap	Mill Mahone Sr.- Makah Fisheries Enhancement	Lambs Thriftway/Marisource
Fish Plate	Rodney Knobel- ODFW	Rangens
Knife Sharpener	Cindy Rathbone- NMFS	Common Sensing
Jacket	Peter Long- USFWS	Christensen Net Works
Sweat Shirt	Gary Yeager- ODFW	Nelson & Sons/ Silver Cup Feeds
Gerber Knife	Paul Preston-Nanaimo River Fish Hatchery	Bioproducts
North Face Pack	John Frost- WDFW	Jensorter
Wine/T-Shirt/Hat	Pete Campbell- DFO- Canada	Lambs Thriftway/Leavenworth Hatchery
Bushnell Binoculars	Jim Bowker- USFWS	Western Chemical
Wine/T-Shirt/Cook Book	Kaare Julshamn-NMFS	Lambs Thriftway/ Sunshine Coast Salmonid Enhancement Society/ Martha Klontz
Fishing Vest	Shelly Peterson- WDFW	Western Chemical

<u>Prize</u>	<u>Recipient</u>	<u>Donor</u>
Gerber Knife	Randy Boise- Warm Springs	Bioproducts
Dip Net and Hat	Glen Rasmusson- DFO- Canada	Edgar Inc./Marisource
2 Bottles of Wine and T- Shirt	Rich Bryant- CDFG	Lambs Thriftway/Rangens
Fish Plate	Steve Pasteur- USFWS	Rangens
Sweatshirt	C. Ledford- Puget Sound Energy	Leavenworth National Fish Hatchery
Coleman Lantern	Ted Calavan- ODFW	Common Sensing
Leatherman Micro Tool	Richard Colvin- WDFW	Common Sensing
Meal Package	Doreen Ross- WDFW	Intermountain Weighing
Kathryn Kostow Print	Mitch Daniel- Nez Perce	Kathryn Kostow
Coleman Lantern	Dick Ewing- Biotech	Common Sensing
2 Bottles of Wine and a Hat	Sam Woods- ODFW	Lambs Thriftway and Marisource
Aladdin Thermos	Dave Owsley- USFWS	Intermountain Weighing Inc.
Fish Print/ T-Shirt	Seth Cooney- ODFW	Western Chemical/ Leavenworth NFH
Kathryn Kostow Print	Eric Wagner- Utah Division of Wildlife Resources	Kathryn Kostow
Gerber Knife and T-Shirt	Don Farmer- Puget Sound Energy	Bioproducts/Leavenworth NFH
Fish Print/ Sweatshirt	Tony Amandi- ODFW	Western Chemical/ Leavenworth NFH
Maglight Flashlight	Daniel Free- USFWS	Common Sensing

<u>Prize</u>	<u>Recipient</u>	<u>Donor</u>
2 Bottles of Wine	Tom Kane- USFWS	Lambs Thriftway
2 Bottles of Wine	Brian Lyon- WDFW	Lambs Thriftway
Christmas Ornament/Cap	Dan Dunn- CEDC Fisheries	VMG Industries/Marisource
Fish Plate	Bob Becker- ODFW	Rangens
Buck Knife	Susan Stanley	Bioproducts
Fishing Pole/Reel/Cap	Mike Finklin- Puget Sound Energy	Marisource
Dutch Oven	Lincoln Feddersen-Colville Tribe	NWFCC
Salishan Vacation Package	Dan Davies- USFWS	Salishan Lodge
Salishan Vacation Package	Greg Lipsiea- ODFW	Salishan Lodge

# NORTHWEST FISH CULTURE CONFERENCE HISTORICAL RECORD

<u>YEAR</u>	<u>LOCATION</u>	<u>HOST AGENCY</u>	<u>CHAIRMAN</u>
1950	PORTLAND, OR	U.S. FISH AND WILDLIFE SERVICE	TED PERRY
1951	WENATCHEE, WA	U.S. FISH AND WILDLIFE SERVICE	ROGER BURROWS
1952	SEATTLE, WA	WASHINGTON DEPT. OF FISHERIES	BUD ELLIS
1953	PORTLAND, OR	FISH COMMISSION OF OREGON	FRED CLEAVER
1954	SEATTLE, WA	U.S. FISH AND WILDLIFE SERVICE	BOB RUCKER
1955	PORTLAND, OR	OREGON GAME COMMISSION	JOHN RAYNER
1956	SEATTLE, WA	WASHINGTON DEPT. OF GAME	CLIFF MILLENBACH
1957	PORTLAND, OR	U.S. FISH AND WILDLIFE SERVICE	HARLAN JOHNSON
1958	SEATTLE, WA	WASHINGTON DEPT. OF FISHERIES	BUD ELLIS
1959	PORTLAND, OR	FISH COMMISSION OF OREGON	ERNE JEFFRIES
1960	OLYMPIA, WA	WASHINGTON DEPT. OF GAME	JOHN JOHANSEN
1961	PORTLAND, OR	OREGON GAME COMMISSION	CHRIS JENSEN
1962	LONGVIEW, WA	U.S. FISH AND WILDLIFE SERVICE	ROGER BURROWS
1963	OLYMPIA, WA	WASHINGTON DEPT. OF FISHERIES	BUD ELLIS
1964	CORVALLIS, OR	OREGON STATE UNIVERSITY	JOHN FRYER
1965	PORTLAND, OR	U.S. FISH AND WILDLIFE SERVICE	JOHN HALVER
1966	PORTLAND, OR	FISH COMMISSION OF OREGON	WALLY HUBLON
1967	SEATTLE, WA	UNIVERSITY OF WASHINGTON	LOREN DONALDSON
1968	BOISE, ID	IDAHO DEPT. OF FISH AND GAME	PAUL CUPLIN
1969	OLYMPIA, WA	WASHINGTON DEPT. OF GAME	JOHN JOHANSEN
1970	PORTLAND, OR	OREGON GAME COMMISSION	CHRIS JENSEN
1971	PORTLAND, OR	U.S. FISH AND WILDLIFE SERVICE	MARV SMITH
1972	SEATTLE, WA	WASHINGTON DEPT. OF FISHERIES	DICK NOBLE
1973	WEMME, OR	OREGON FISH COMMISSION	ERNE JEFFRIES

<u>YEAR</u>	<u>LOCATION</u>	<u>HOST AGENCY</u>	<u>CHAIRMAN</u>
1974	SEATTLE, WA	UNIVERSITY OF WASHINGTON	ERNIE SALO
1975	OTTER CREST, OR	OREGON STATE UNIVERSITY	JACK DONALDSON
1976	TWIN FALLS, ID	UNIVERSITY OF IDAHO	BILL KLONTZ
1977	OLYMPIA, WA	WASHINGTON DEPT. OF GAME	JIM MORROW
1978	VANCOUVER, WA	U.S. FISH AND WILDLIFE SERVICE	DAVE LEITH
1979	PORTLAND, OR	OREGON DEPT. OF FISH AND WILDLIFE	ERNIE JEFFRIES
1980	COURTENAY, B.C.	FISHERIES & OCEANS, CANADA	KEITH SANDERCOCK
1981	OLYMPIA, WA	WASHINGTON DEPT. OF FISHERIES	WILL ASHCRAFT
1982	GLENEDEN BEACH, OR	NATIONAL MARINE FISHERIES SERVICE	EINAR WOLD
1983	MOSCOW, ID	UNIVERSITY OF IDAHO & IDAHO DEPT. OF FISH AND GAME	BILL KLONTZ & EVAN PARRISH
1984	KENNEWICK, WA	WASHINGTON DEPT. OF GAME	JIM GEARHEARD
1985	TACOMA, WA	U.S. FISH AND WILDLIFE SERVICE	ED FORNER
1986	EUGENE, OR	OREGON DEPT. OF FISH AND WILDLIFE	CHRIS CHRISTENSEN
1987	TACOMA, WA	WASHINGTON DEPT. OF FISHERIES	WILL ASHCRAFT
1988	RICHMOND, B.C.	B.C. MINISTRY OF ENVIRONMENT	DON PETERSON & PETER BROWN
1989	GLENEDEN BEACH, OR	NATIONAL MARINE FISHERIES SERVICE	RZ SMITH
1990	BOISE, ID	IDAHO DEPT. OF FISH AND GAME	BILL HUTCHINSON
1991	REDDING, CA	CALIFORNIA DEPT. OF FISH AND GAME	KEN HASHAGEN
1992	WENATCHEE, WA	WASHINGTON DEPT. OF WILDLIFE & ALASKA DEPT. OF FISH AND GAME	JOHN KERWIN & IRV BROCK
1993	SPOKANE, WA	U.S. FISH AND WILDLIFE SERVICE	ED FORNER
1994	SUNRIVER, OR	OREGON DEPT. OF FISH AND WILDLIFE	RICH BERRY
1995	FIFE, WA	WASHINGTON DEPT. OF FISH AND WILDLIFE	LARRY PECK
1996	VICTORIA, B.C.	B.C. MINISTRY OF ENVIRONMENT, LANDS, AND PARKS & DEPT. OF FISHERIES AND OCEANS CANADA	DON PETERSON & GREG BONNELL

<u>YEAR</u>	<u>LOCATION</u>	<u>HOST AGENCY</u>	<u>CHAIRMAN</u>
1997	GLENEDEN BEACH, OR	NATIONAL MARINE FISHERIES SERVICE	RZ SMITH
1998	BOISE, ID	IDAHO DEPARTMENT OF FISH AND GAME	TOM ROGERS
1999	TO BE ANNOUNCED	U.S. FISH AND WILDLIFE SERVICE	TO BE ANNOUNCED