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PROCEEDINGS OF THE
FORTIETH ANNUAL
NORTHWEST FISH CULTURE
CONFERENCE



GLENEDEN BEACH, OREGON

DECEMBER 5 - 7, 1989

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OF THE
FORTIETH ANNUAL
NORTHWEST FISH
CULTURE CONFERENCE

DECEMBER 5 - 7, 1989

GLENEDEN BEACH, OREGON



**COLUMBIA RIVER
INTER-TRIBAL
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NATIONAL MARINE FISHERIES SERVICE

THE NORTHWEST FISH CULTURE CONFERENCE

Northwest Fish Culture Conferences are annual, informal meetings held for the 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 developments, and research studies are presented. Both within the meeting and outside the formal 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. The subject matter is limited to topics that have direct application to fish culture.

These PROCEEDINGS contain unedited briefs of oral reports presented at the conference. Much of the material involves progress of uncompleted studies or reports. THESE INFORMAL RECORDS SHOULD NOT BE INTERPRETED OR QUOTED AS A PUBLICATION.

Mention in these PROCEEDINGS does not indicate approval, recommendation, or endorsement of any proprietary product or propriety material.

PREFACE

Over 375 enthusiastic people with widely varied backgrounds but with a common interest in fish husbandry attended the Fortieth Annual Northwest Fish Culture Conference. Given the short supply of travel funds experienced by most fisheries agencies, I feel that this represents a significant commitment of time and funds and shows the importance that is placed on the value of this conference. In addition to being treated to unparalleled meeting facilities and meals, those attending the conference spent three days being exposed to "state-of-the-art" presentations covering topics ranging from hatchery orientations and hatchery practices, to discussions covering the use of the drug "Erythromycin" across fisheries agencies in the Region. As most who attend these meetings, the valuable information received in the meeting room is often matched or exceeded by the information exchanged and the contacts made during breaks, in the halls, or after hours.

Commercial representatives provided exhibits of fish culture equipment and technology. They graciously hosted the coffee breaks. This was very much appreciated and significantly added to the opportunities for personal contacts during the meeting. These representatives were very generous in providing prizes for the drawings held throughout the conference. I would like to give a special thanks to the four fish feed companies, Silver Cup Feeds, Rangen Feeds, Moore Clark Company, and Bioproducts Inc., for hosting the outstanding hospitality hour.

Planning and carrying out a conference such as this one is a monumental undertaking. Without the support and able assistance I received Mike Delarm, it couldn't have been done. In addition to helping with the program and registration, Mike single handedly took care of all the arrangements for the commercial exhibits and the prize drawings. This is a thankless task, especially where solicitation of prizes is concerned, but is an absolute necessary.

In addition to Mike, several others from the Environmental and Technical Services Division of the National Marine Fisheries Service were instrumental in the functioning of the conference. Mattie Crosby and Barbette Falk provided assistance for registration. Mike and Robert Vreeland served as Session Leaders. Randy Lee, provided general support and was responsible for seeing that the audio/visual equipment functioned. Randy was also drafted to help with the drawings. Support was provided from outside this office by Ron Morinaka of the Bonneville Power Administration who graciously volunteered to serve as Session Leader for two sessions. I would like to acknowledge these people and thank them for their tireless support.

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 group as large as this to give a presentation. It represents a significant commitment of time and effort. Feedback I have received on the presentations has been nothing but positive and I think that all of these people should receive commendations for their presentations.

I have enjoyed working on this project and I am looking forward to the 1990 meeting in Boise, Idaho, December 4-6.

R. Z. Smith

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REPORT ON THE VOLITIONAL RELEASE OF THREE BROODS OF COHO SALMON

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Washington State Department
of Fisheries

ABSTRACT

Three broods of coho salmon (*Oncorhynchus kisutch*) at four hatcheries were involved in an experiment to determine what, if any, advantages were available if coho salmon are released volitionally rather than forced out of the ponds (flushed release).

Two hatcheries had volitional and control groups of coded wire tagged fish to compare overall survivals, size of returning adults, and changes in ocean distribution. The remaining two hatcheries released fish from large rearing vessels with populations exceeding one million fish.

Electronic fish counters were installed in the volitional release ponds and numbers of fish which had migrated were recorded twice every 24 hours.

Survival of volitionally released fish was equal to or greater than the control groups in most cases. Size of adults from volitional groups was equal to or exceeded the size from control groups in every case. The percentage of jacks (precocious males) was greater for the volitional groups than the control groups in every case. Ocean distribution appeared similar for both groups. Advantages for hatchery operation include; reduction of loadings at critical times, increased flexibility for hatchery managers in using available water. Other advantages include release of smolts which may decrease intra- and inter specific competition.

INTRODUCTION

The seaward migration of juveniles is a commonly known behavior trait of Pacific salmon.

This event has taken place annually for ten's of thousands of years in naturally spawning population. It is known that this takes place spontaneously within individuals and the time is variable. Age of the individual fish, growth rate, water temperatures, photoperiod, physiological development, and genetics are only some of the factors which play a role in determining the exact time any individual fish begins this journey.

From an evolutionary standpoint, this seems to make sense. In most natural systems, diversity fosters stability, and stability is one method of ensuring the longevity of a species.

When salmonids are placed in a hatchery environment for part of their life cycle, some compromises are usually made. Because fish culturists must stay within the basic biological requirements of a particular species, they have been somewhat limited in the amount of manipulation which can be done while operation a production sized salmon hatchery.

One common practice which has been carried out for many years is the simultaneous release of all fish from an individual hatchery pond. Usually, at some pre-selected time and/or average size, the screen is pulled and the pond drained.

This practice has many advantages for the fish culturist; planning for water use, pond space requirements, and food purchases can be made well in advance when the rearing period is fixed. Because of the vast amount of research concerning the optimum time and size to release fish, the culturist can select a time and size within the known range and expect to meet with some success.

Despite this, the idea of allowing fish to migrate volitionally from a hatchery seems, intuitively, like the right thing to

do to some culturists and biologists. The idea that volitional release may increase survival, ease pond loading at critical times, or reduce intra- or interspecific competition for downstream migrants is incentive enough to cause the culturist to ponder this strategy.

A major obstacle in evaluation volitional release techniques is the enumeration of outmigrants. By knowing the number of fish that have migrated, the culturist can adjust feed or disease treatments and regulate water requirements when necessary. Biologists also require an enumeration techniques so aspects of fish behavior and physiology during release can be monitored.

The Washington Department of Fisheries has been actively involved in evaluating electronic fish counting hardware for several years. These fish counters have provided a tool to assess volitional release strategies.

METHODS

Four hatcheries were chosen for participation in this study; Grays River, Kalama Falls, Elokomin, and Klickitat. Elokomin and Klickitat were chosen because they have single large release ponds containing 1 million or more fish. Grays River and Kalama Falls were used because coho are normally released from several identical ponds, thus providing a control group for survival comparisons. The control ponds at Grays River and Kalama Falls were released on a specific day according to program requirements. The experimental ponds at Grays River and Kalama Falls and the large release ponds at Elokomin and Klickitat had a FC-3 electronic fish counter (Northwest Marine Technology) installed in April and fish were allowed to leave volitionally. The volitional release ponds at all stations were set up so fish could return to the pond after passing through the counter. Since the counters have the capability to count upstream as well as downstream passage and compute the difference, accurate counts were recorded. The design of the outlet structures provided a small forebay for fish to hold in before moving over dam boards or into the sump and down the pond drain. This procedure

helped to ensure the fish were leaving volitionally. Counts of less than 50 fish per day moving back into the pond were common and support the notion that fish which passed through the counter were showing a desire to move downstream.

Coho at all facilities were spawned, incubated, hatched, tagged, and reared using standard practices for those hatcheries. The study was repeated at each station using the 1983, 84, and 85 brood coho.

In order to ensure the marked fish were truly representing the entire population, mark rates approaching 100% were used at the control stations (Grays River and Kalama Falls). Since Elokomin and Klickitat had ponds with over 1 million fish, high mark rates were not practical; therefore, an attempt was made to sample outmigrants throughout the release for the presence of AD+CWT fish to ensure that marked and unmarked fish were behaving similarly.

Numbers of outmigrants were recorded at each facility at 8:00 AM and 4:30 PM daily. The 4:30 PM total was considered the "day count" and the 8:00 AM total was considered the "night count".

To determine size differences between outmigrants and fish residing in the pond, length samples were taken periodically throughout the outmigration period. The samples were taken at Elokomin for the 1983 and 1985 broods and at Klickitat for the 1984 and 1985 broods.

At Grays River and Kalama Falls, the volitional outmigrants were sampled on several days to determine average size at release. Control ponds were sampled on the day of release.

RESULTS

The fish behavior observations recorded in this study have been previously reported at other Fish Culture Conferences. The discussion therefore will be brief and limited to general principles which made themselves evident during the study. This report will focus on the coded-wire tag recoveries of the adults produced from the various study groups.

FISH BEHAVIOR:

The average release duration at all four hatcheries correlates with the number of fish in the pond. Grays River had the fewest fish and the shortest duration (ave. =32 days), followed by Kalama Falls (ave. =42 days), Elokommin (ave. =52 days), and Klickitat (ave. =66 days).

The highest number of outmigrants for a twenty-four hour period was 110,000 fish which occurred at Elokommin on May 25, 1983. There were numerous days at either Klickitat or Elokommin which had 50,000 or more fish migrating (Fig. 1).

The averages at each hatchery across brood years show a higher per-hour rate of migration during the 8 hour "day" than the 16 hour "night". However, while the "day" rate was higher, most of the fish (% of the total) migrated during the "night" (Fig. 2).

The percent of fish which volitionally migrated was very consistent at all stations and across all brood years. An average of 99%+ at Klickitat, 98% at Elokommin, 96% at Kalama Falls, and 92% at Grays River demonstrated clearly that yearling coho at these facilities can and will negotiate a variety of outfall structure successfully. The 92% overall average at Grays River was an artifact of pond space requirements, which forced the termination of the volitional release of the 1985 brood after only 83% of the fish had migrated (Fig. 3 and 4). Sampling for average size of outmigrants was a difficult chore as movements of fish could not be precisely foreseen. For all but two sampling dates, outmigrants were significantly larger than fish which remained in the pond ($p < 0.05$) when tested using ANOVA. The coefficient of variation was also lower on the outmigrant samples, indicating less variation around the mean length.

PRELIMINARY AD+CWT RECOVERIES:

GRAYS RIVER:

The 1983 brood volitional release group had a significant increase ($p < 0.05$) in survival over the control group (3.44% vs 2.33%) respectively. The 1984 and 1985

brood volitional release groups had slightly lower survivals, although not significantly, than the respective control groups. The three year average of volitional vs control survivals was 2.18% vs 1.85% respectively (Table 1; Fig. 5).

The adults from the volitional release groups were larger in every case than the adults from the control groups (648 mm vs 633 mm average for three years). Statistical significance has not been tested but no overlap in the value of the replicates was recorded (Fig. 6).

The volitional release groups produced a higher percentage of jacks than the control groups in every brood year. The average over the three brood years was 5.4% vs 3.67% jacks for the volitional and control groups respectively (Fig. 7).

KALAMA FALLS:

The 1983 brood volitional release group had a significant increase ($p < 0.05$) in survival over the control group (7.71% vs 6.1%) respectively. The 1984 brood volitional release groups had significantly ($p < 0.05$) lower survival than the control group (0.56% vs 1.65% respectively). The 1985 brood volitional and control groups survivals were not significantly different (6.33% vs 6.63%). The three year average of volitional vs control survivals was 4.87% vs 4.79% respectively (Table 2; Fig. 8).

The adults from the volitional release groups were larger in every case than the adults from the control groups (633 mm vs 604 mm average for three years). Statistical significance has not been tested but no overlap in the value of the replicates was recorded (Fig. 9).

The volitional release groups produced a higher percentage of jacks than the control groups in every brood year. The average over the three brood years was 0.78% vs 0.47% jacks for the volitional and control groups respectively (Fig. 10).

ELOKOMIN AND KLICKITAT:

No control groups were available for either of these hatcheries. Their primary role in this study was to allow the observation of

smolting behavior in large populations of juveniles over a protracted period of time. Survival data from volitional release groups will need to be compared with historical data before conclusions can be drawn.

strategy. Overall survival is comparable with normal release practices, the level of pond loading is reduced before it becomes critical at most facilities, adult size can be increased and more flexibility in the use of available water is gained by the hatchery manager.

CONCLUSIONS

The volitional release of coho smolts from production hatcheries can be a useful

TABLE 1. PRELIMINARY RESULTS OF VOLITIONAL RELEASE STUDIES
(10/10/89)

GRAYS RIVER 1983 BROOD COHO

| TAG CODE | NUMBER RELEASE | STUDY PURPOSE | NUMBER JACKS | NUMBER ADULTS | TOTAL % SUR | AVE. SUR. | AVE. ADULT FK LENGTH |
|----------|----------------|---------------|--------------|---------------|-------------|-----------|----------------------|
| 633259 | 24678 | VOL. | 32 | 916 | 3.85 | | 625 (mm) |
| 633260 | 24274 | VOL. | 23 | 222 | 3.44 | 3.44 | 624 (mm) |
| 633261 | 24695 | VOL. | 42 | 708 | 3.03 | | 624 (mm) |
| 633262 | 24599 | CONT. | 10 | 533 | 2.20 | | 606 (mm) |
| 633263 | 24579 | CONT. | 11 | 513 | 2.14 | 2.33 | 613 (mm) |
| 633301 | 23941 | CONT. | 9 | 626 | 2.65 | | 607 (mm) |

GRAYS RIVER 1984 BROOD COHO

| TAG CODE | NUMBER RELEASE | STUDY PURPOSE | NUMBER JACKS | NUMBER ADULTS | TOTAL % SUR | AVE. SUR. | AVE. ADULT FK LENGTH |
|----------|----------------|---------------|--------------|---------------|-------------|-----------|----------------------|
| 633531 | 25851 | VOL. | 4 | 143 | 0.56 | | 674 (mm) |
| 633532 | 26277 | VOL. | 1 | 111 | 0.42 | 0.49 | 667 (mm) |
| 633533 | 26416 | VOL. | 3 | 127 | 0.49 | | 667 (mm) |
| 633534 | 26300 | CONT. | 2 | 106 | 0.41 | | 640 (mm) |
| 633535 | 25853 | CONT. | 0 | 160 | 0.61 | 0.54 | 649 (mm) |
| 633536 | 26191 | CONT. | 0 | 158 | 0.60 | | 666 (mm) |

GRAYS RIVER 1985 BROOD COHO

| TAG CODE | NUMBER RELEASE | STUDY PURPOSE | NUMBER JACKS | NUMBER ADULTS | TOTAL % SUR | AVE. SUR. | AVE. ADULT FK LENGTH |
|----------|----------------|---------------|--------------|---------------|-------------|-----------|----------------------|
| 634247R6 | 39541 | VOL. | 92 | 1014 | 2.79 | | 654 (mm) |
| 634249R6 | 39231 | VOL. | 82 | 1054 | 2.89 | 2.84 | 652 (mm) |
| 634250R6 | 39874 | CONT. | 72 | 1101 | 2.94 | | 642 (mm) |
| 634252R6 | 39142 | CONT. | 65 | 1233 | 3.31 | 3.12 | 642 (mm) |

TABLE 2. PRELIMINARY RESULTS OF VOLITIONAL RELEASE STUDIES
(10/10/89)

KALAMA FALLS 1983 BROOD COHO

| TAG CODE | NUMBER RELEASE | STUDY PURPOSE | NUMBER JACKS | NUMBER ADULTS | TOTAL % SUR | AVE. SUR. | AVE. ADULT FK. LENGTH |
|----------|----------------|---------------|--------------|---------------|-------------|-----------|-----------------------|
| 633156 | 51214 | VOL. | 34 | 3887 | 7.65 | | 605 (mm) |
| 633157 | 51340 | VOL. | 13 | 3983 | 7.78 | 7.71 | 604 (mm) |
| 633232 | 50886 | CONT. | 8 | 3179 | 6.26 | | 603 (mm) |
| 633233 | 51014 | CONT. | 7 | 3024 | 5.94 | 6.10 | 601 (mm) |

KALAMA FALLS 1984 BROOD COHO

| TAG CODE | NUMBER RELEASE | STUDY PURPOSE | NUMBER JACKS | NUMBER ADULTS | TOTAL % SUR | AVE. SUR. | AVE. ADULT FK. LENGTH |
|----------|----------------|---------------|--------------|---------------|-------------|-----------|-----------------------|
| 633454 | 46889 | VOL. | 5 | 306 | 0.66 | | 643 (mm) |
| 633455 | 47358 | VOL. | 3 | 224 | 0.47 | 0.56 | 654 (mm) |
| 633456 | 52823 | CONT. | 11 | 828 | 1.58 | | 613 (mm) |
| 633457 | 52544 | CONT. | 5 | 899 | 1.72 | 1.65 | 613 (mm) |

KALAMA FALLS 1985 BROOD COHO

| TAG CODE | NUMBER RELEASE | STUDY PURPOSE | NUMBER JACKS | NUMBER ADULTS | TOTAL % SUR | AVE. SUR. | AVE. ADULT FK. LENGTH |
|----------|----------------|---------------|--------------|---------------|-------------|-----------|-----------------------|
| 634216R6 | 38976 | VOL. | 18 | 2486 | 6.42 | | 664 (mm) |
| 634219R6 | 39194 | VOL. | 32 | 2419 | 6.25 | 6.33 | 668 (mm) |
| 634221R6 | 38919 | CONT. | 15 | 2580 | 6.66 | | 650 (mm) |
| 634222R6 | 38964 | CONT. | 17 | 2557 | 6.60 | 6.63 | 649 (mm) |

FIGURE 1

ELOKOMIN 1983,84,85 BROOD COHO OUTMIGRATION TOTALS

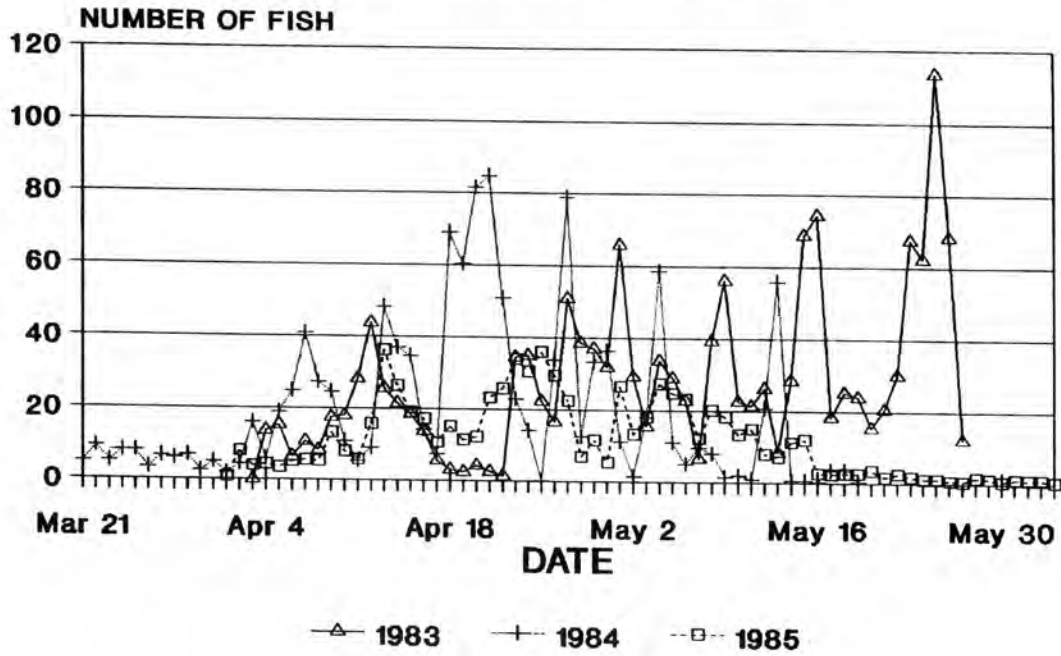


FIGURE 2

KLICKITAT 1985 BROOD COHO DAY VS NIGHT

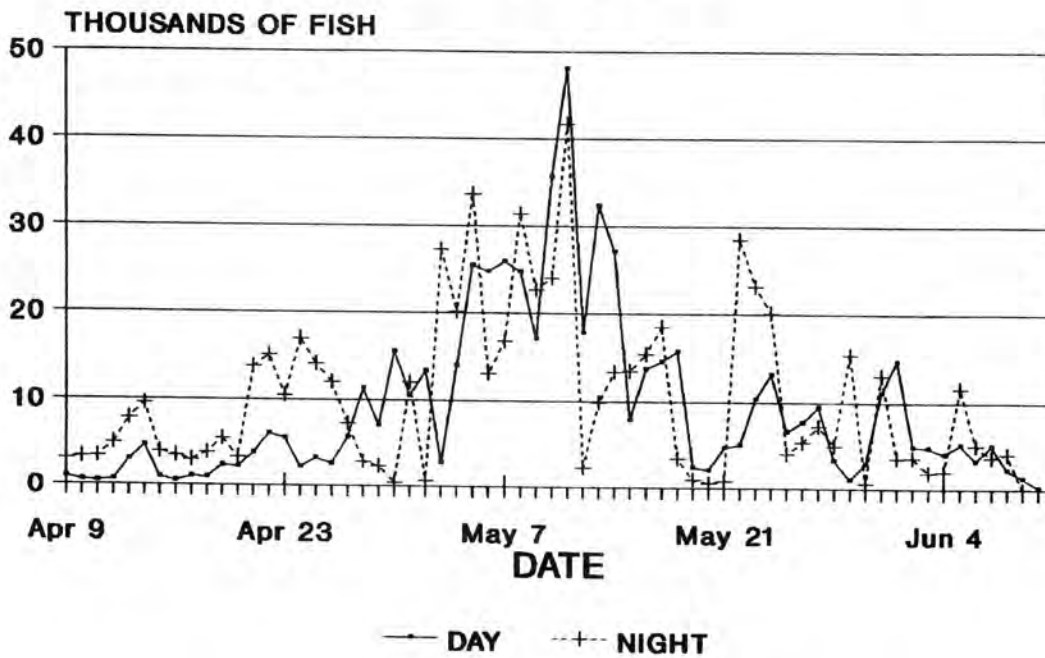


FIGURE 3

GRAYS RIVER 1983,84,85 BROOD COHO OUTMIGRATION PERCENT

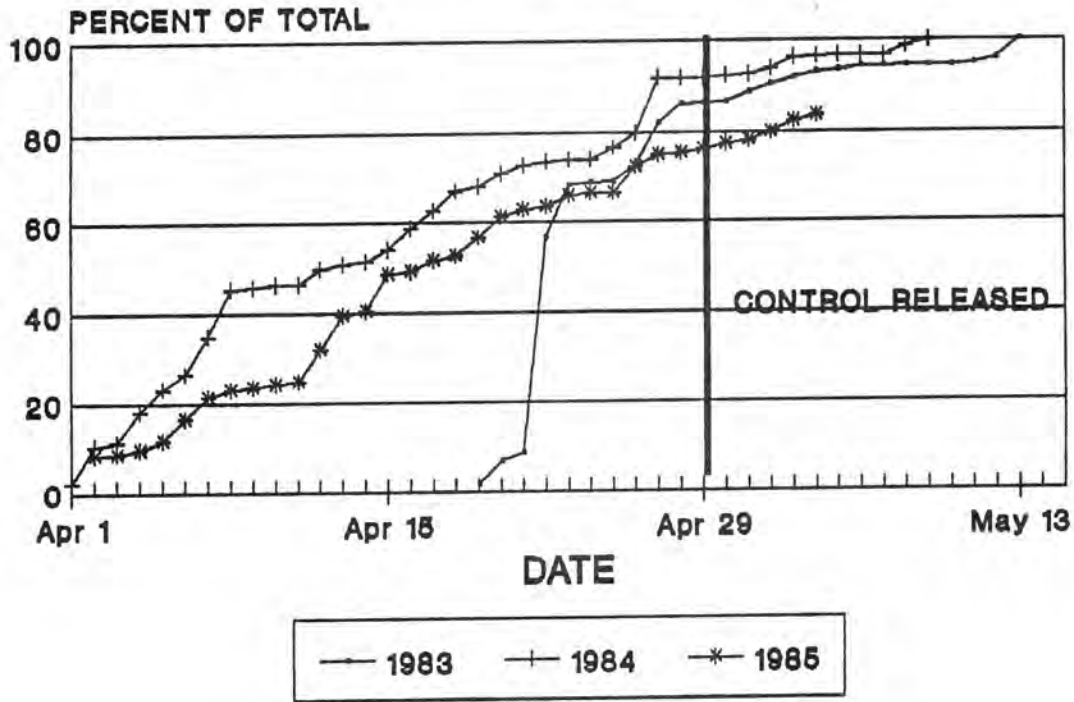


FIGURE 4

KALAMA FALLS 1983,84,85 BROOD COHO VOLITIONAL OUTMIGRATION PERCENT

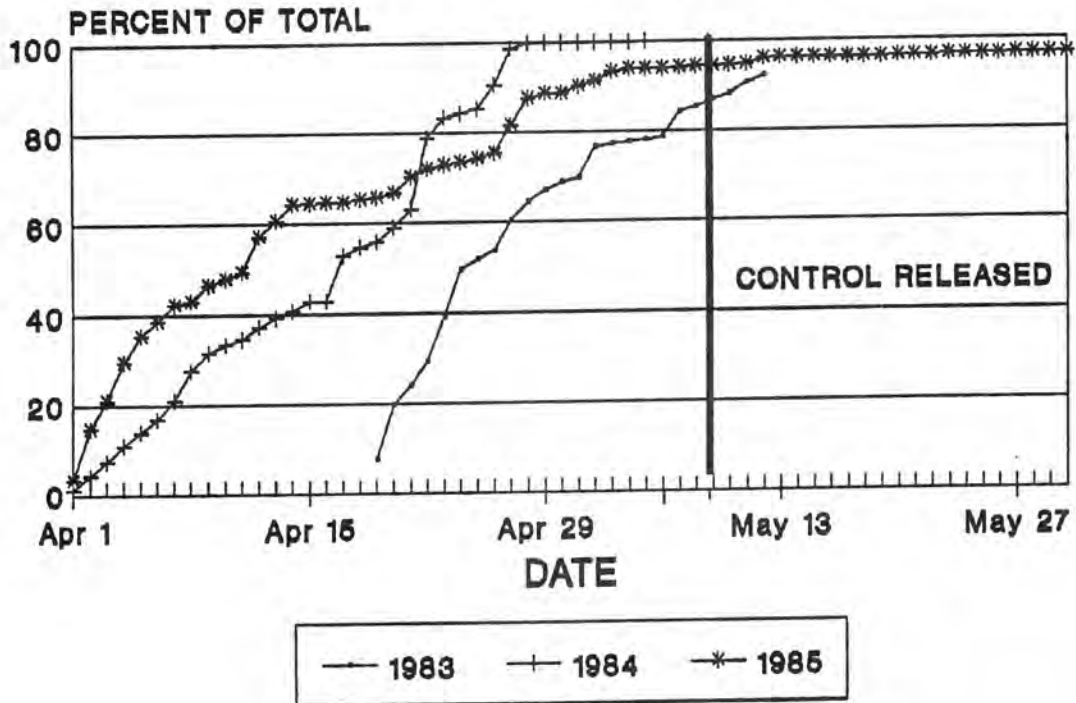


FIGURE 5

SURVIVAL OF VOLITIONALLY RELEASED COHO GRAYS RIVER HATCHERY 1983-1985 BROODS

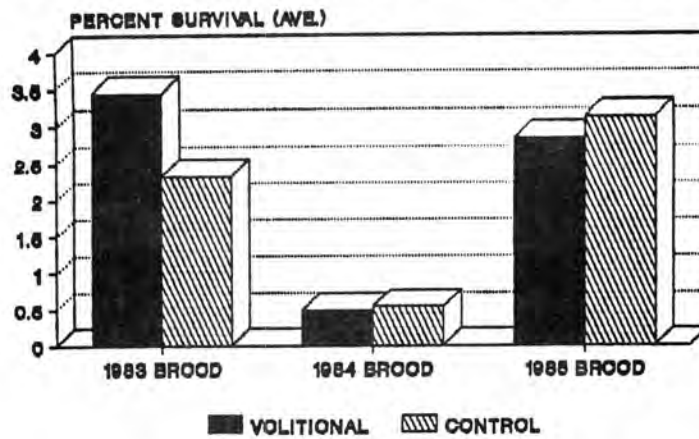


FIGURE 6

SIZE OF ADULTS VS. TYPE OF RELEASE GRAYS RIVER HATCHERY 1983-1985 BROODS

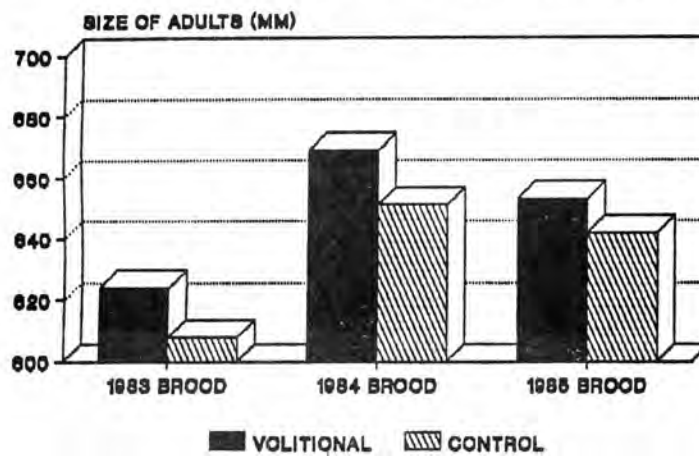


FIGURE 7

GRAYS RI. JACK PRODUCTION BASED ON TYPE OF RELEASE

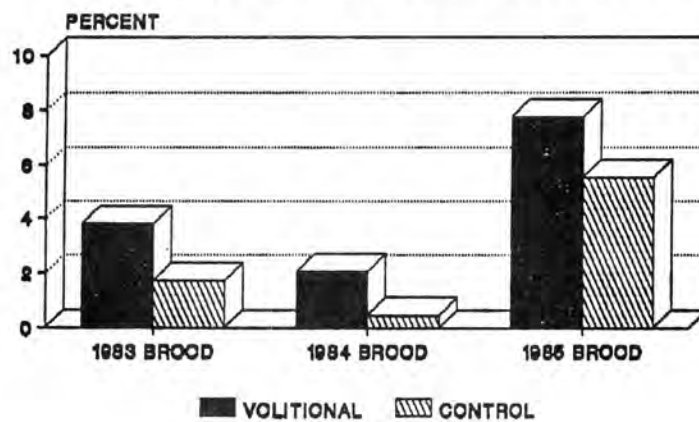


FIGURE 8

SURVIVAL OF VOLITIONALLY RELEASED COHO KALAMA FALLS HATCHERY 1983-1985 BROODS

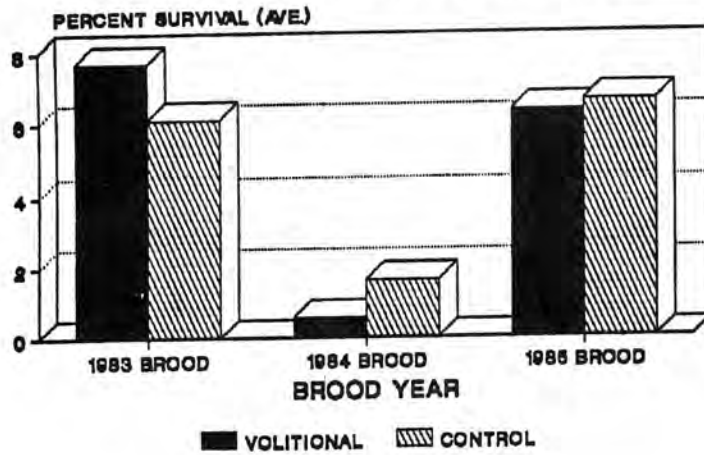


FIGURE 9

SIZE OF ADULTS VS TYPE OF RELEASE KALAMA RIVER HATCHERY 1983-1985 BROODS

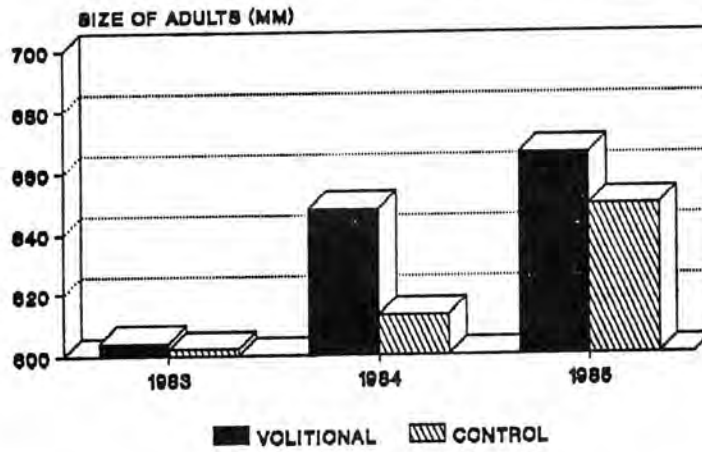
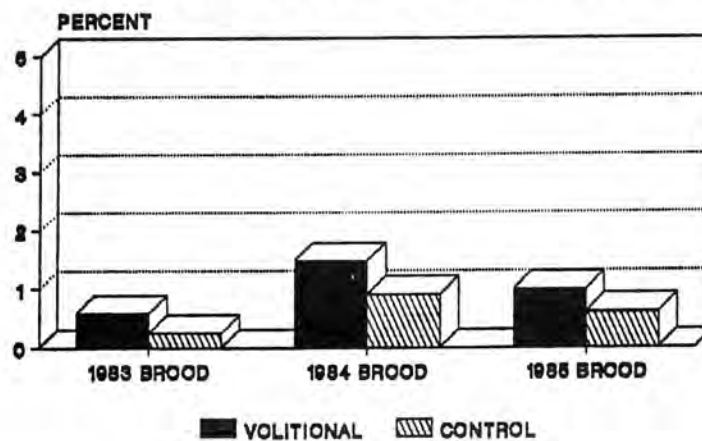


FIGURE 10

KALAMA FALLS JACK PRODUCTION BASED ON TYPE OF RELEASE



**EQUIPMENT FOR SIPHON RELEASE OF '86 AND '87 COHO
AT WASHOUGAL SALMON HATCHERY**

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**Washington Department Fisheries
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This hatchery's design does not allow for the volitional release of fish from its raceways. To compensate for this, a siphon was implemented.

The siphon consisted of a three inch diameter type-w suction water hose, with a fabricated ten inch diameter tunnel at the head end. The siphon was started using a venturi. The hose was then placed in the outfall vault to achieve a head difference of approximately fifty-five inches. Our intent was to maintain a velocity in the hose less than the burst swimming speed of the fish to prevent drawing fish out of the pond. The water velocity was forty-two inches per second (determined with a ping pong ball, a watch, and a known length). The discharge was seventy-six gallons per minute.

The first season, we used two hoses, one per pond, and switched ponds every two days. Our count samples began late April. The counts were taken three times daily for a five minute interval. The samples ranged from zero to three hundred fifty for each period. The coho leaving averaged nineteen fish per pound throughout the month the siphons were operating. Those coho remaining in the pond averaged twenty fish per pound. It

was estimated that fifty percent of the half million coho planted were effectively released via the siphon method by late May.

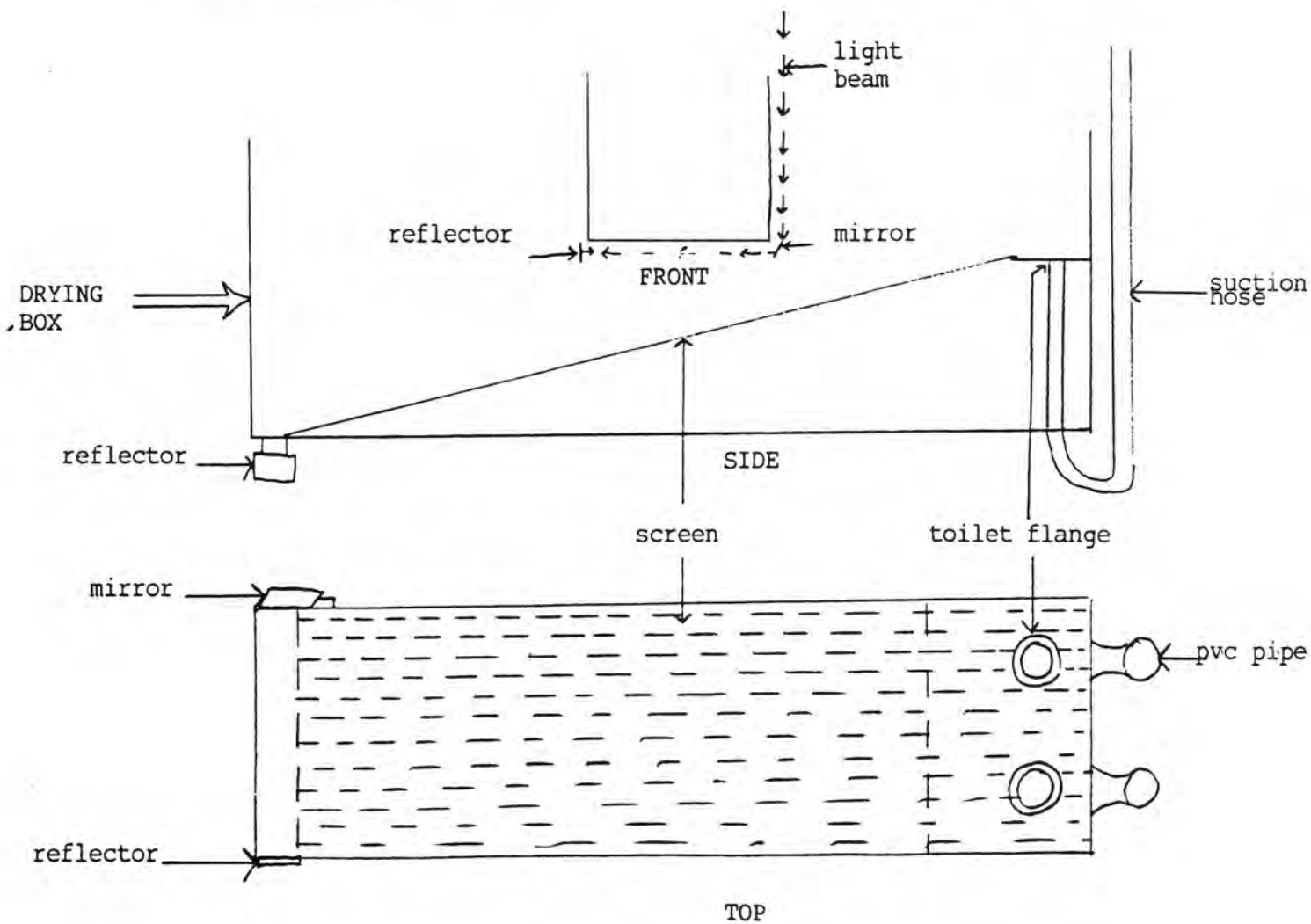
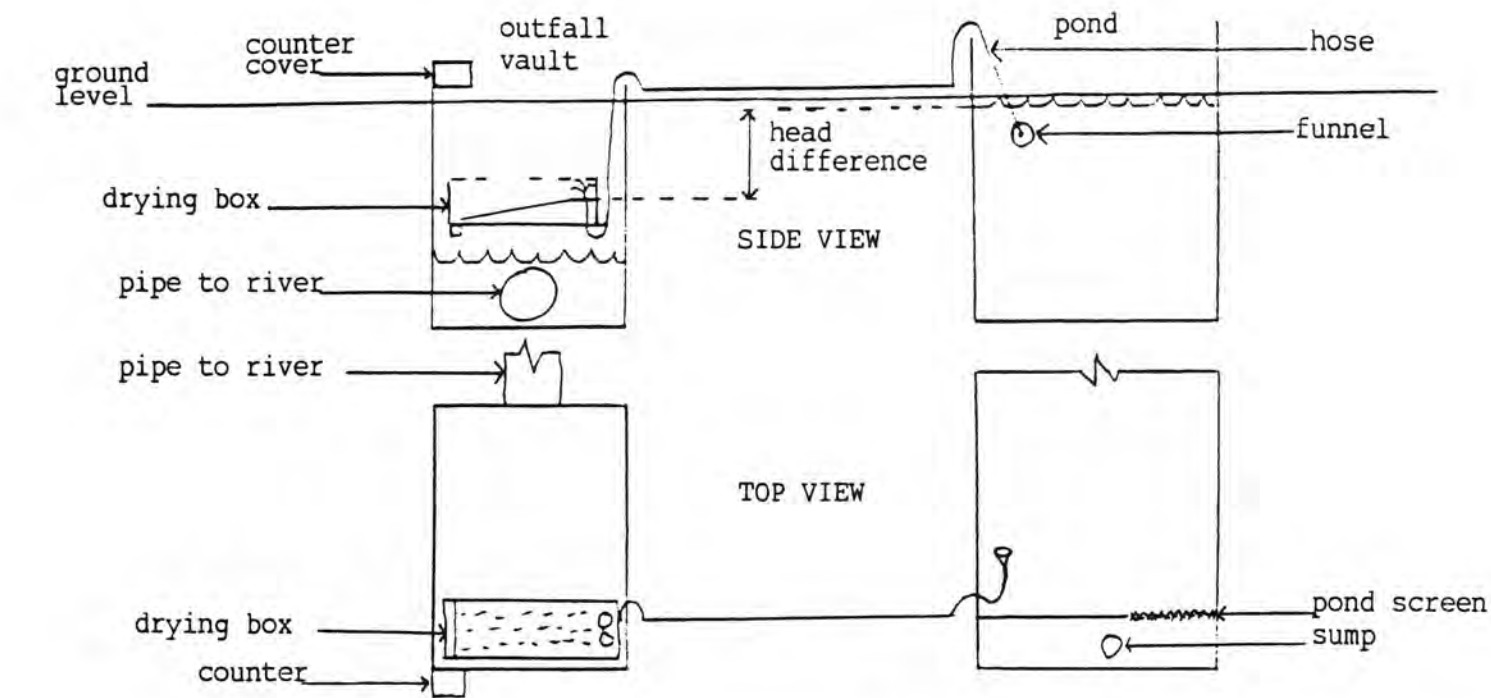
The second season we incorporated a drying box attached to the tail end of the siphon hose. The drying box was constructed of a metal frame and wooden sides. An upwell delivered the coho from the siphon to a slotted screen. The fish gently rolled down the screen, passed through a slot, breaking a light beam which triggered an automatic counter. Our purpose was to keep track of the coho release through out the day. With the counter, we were able to have more accurate numbers over a longer period of time.

Water and space were our main concerns at this time of the year, as at so many other hatcheries, Water temperatures were rising, more space was needed for splits, and densities for yearlings were approaching maximum. The siphons helped reduce the pond loadings and at the same time we were able to use water elsewhere.

If you decide to release fish volitionally, the siphon method works efficiently and effectively.

MATERIALS LIST

| | |
|-----------------------------|---------------------|
| Aluminum Pond Screen..... | 5/32" X 1" Slot |
| Electronic Counter..... | 7600 Series, Grange |
| IR Photo Alarm..... | Radio Shack #49307 |
| Toilet Flange and Trap..... | Local Plumbing |
| Type-W 3" Suction Hose..... | Hatchery Supplies |
| 1/2" Plywood..... | Local Lumber |
| 1" Angle Iron..... | Hatchery Supplies |



SCHEMATIC

**SURVIVAL, CONTRIBUTION, AND RETURN OF HATCHERY
COHO SALMON RELEASED INTO FRESHWATER,
ESTUARINE, AND MARINE ENVIRONMENTS**

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ABSTRACT

We release six groups of marked (coded-wire tagged and adipose fin clipped) yearling hatchery coho salmon *Oncorhynchus kisutch* each year for five years in six locations beginning with the 1981 brood. Fish were released at Bonneville Hatchery (control), at the Tongue Point Coast Guard Station (head of tidewater in the Columbia River), between the jetties at the Columbia River bar, in the Columbia River plume water, in coastal water approximately 12 miles north and 12 miles offshore of the mouth of the river, and in oceanic water approximately 24 miles offshore. We found that the average survival for the fish released at

Tongue Point was 1.6 times the control and that these fish contributed to the inriver gillnet fishery, after adjusting for differences in ocean survival between the groups, at a rate of 2.5 times the control. We found no significant difference between survival of the other release groups and survival of the control group. We also found that the percentage of adult fish that returned to locations other than the Columbia Basin increased as the distance the fish were transported offshore increased.

(This study has been submitted for publication in the Canadian Journal of Fisheries and Aquatic Sciences).

ALTERNATIVE RELEASE STRATEGIES FOR FALL CHINOOK AT WDF COLUMBIA RIVER HATCHERIES

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INTRODUCTION

The Washington Department of Fisheries (WDF) along with the U.S. Fish and Wildlife Service (USFWS) and the Oregon Department of Fish and Wildlife (ODFW) participated in a study to determine the survival, distribution, and value of fall chinook raised at Columbia River hatcheries. This study was funded by the Bonneville Power Administration (BPA) and administered by the National Marine Fisheries Service (NMFS) (Vreeland 1985, 1986, 1987). The study consisted of coded wire tagging approximately 5% of all fall chinook production during the 1978-1981 brood years. Approximately 15 million fish were marked during the 4 years; 6.6 million from WDF hatcheries alone.

WDF became concerned when preliminary data for the 1978 brood indicated poor survival of WDF reared fish compared with other agency releases. By 1983, it was clear that survival of fall chinook released at WDF hatcheries was the poorest of the three agencies releasing fish. For example, average survival rates of chinook released for the Spring Creek National Fish Hatchery (NFH) (USFWS) ranged between 0.45-1.3% (1978-1981 broods). In comparison, average survival rates of WDF reared chinook ranged between 0.1-0.5%.

WDF began addressing the low survival rate problem by comparing its program to the other two agencies' programs. It became clear that several differences existed among agencies, but the most obvious one was the difference in size at release and time of release. WDF's program was to release fish at approximately 100 fish per pound (fpp)

(4.5 h) in June. In contrast, both the Spring Creek NFH and ODFW programs released most fish at sizes larger than 80 fpp (>5.6 g; Figure 1), and almost all fish were released prior to June (Figure 2). Also, at this time, data became available from a juvenile capture program (Dawley et. al., 1984) at Jones Beach, approximately 35 miles upstream for the Columbia River mouth. These data indicated that captured migrants originating from WDF hatcheries were 10-50% larger than at release. This suggested that only the largest fish from a given hatchery were migrating and/or surviving to the Jones Beach area or that fish were growing considerably during migration downstream.

In 1981, WDF received funding through the Columbia River Fisheries Development Program (CRFDP) to undertake studies to identify possible causes of low survival rates in hatchery reared fall chinook. These studies were named Quality Improvement Studies at Columbia River hatcheries (QIS), and initially focussed on mortality occurring in the hatchery. In 1982 (1982-1985 broods) these funds were used to assess survival of chinook released at alternative times and sizes compared to previous programs.

Specifically, at Washougal Hatchery, four groups of fish were coded wire tagged and released on four separate dates (Table 1): June, September, October, and November. Approximately 100,000 fish per group were marked in each brood year, with the exception of the 1982 brood (the June releases could not be marked because of disease problems). In addition, a marked group of Grays River Hatchery fish was transferred to Washougal and released in

1985 and 1986 as part of a hatchery effects study.

Grays River Hatchery was added to the study beginning with the 1984 brood. Two groups of 1984 brood fish were released from Grays River (Table 1), a control (normal June release) and a fall group (October release). In 1985, the same groups were released with the addition of two more groups: 1) fish whose rearing was accelerated by transfer to Ringold Hatchery (spring water) and released in April, and 2) a group of fish transferred from Washougal as part of the hatchery effects study. Also, the Elokomin Hatchery was added to the study in 1986 (1985 brood). At Elokomin, a group of accelerated reared fish were released in April as well as a control group (June release).

RESULTS

Washougal:

Survival rates varied among brood years and treatments. Survival rates of the control (June) and September releases were similar, and higher than the October and November groups (Figure 3). Average survival rates of the control and September groups were approximately 1.25%. Average survival rates of the October and November Groups were about 0.5%

Contribution rates to the Washington, and combined Washington and Oregon coastal fisheries varied among groups (Figure 4). The lowest contribution to these two fisheries was from the June and September groups. The highest contribution was from the November group.

Grays River:

Survival rates of the 1984 brood control and fall release were 2.3% and 3.2%, respectively (Figure 5). These survival rates are much higher than survival rates of the 1978-1981 broods (average: 0.2%). Data for the 1985 broods are not complete, but it appears that the fall release (September) survived at a much higher

rate than the June group. The accelerated group appears to have survived at an intermediate rate.

The 1984 fall release group contributed approximately 300% more fish to the Washington coastal fisheries than the June group (Figure 6). Data are not complete for the 1985 brood.

Elokomin:

Data are not complete for the 1985 brood releases from Elokomin. Preliminary indications are that the accelerated groups survived at a much higher rate and contributed more fish to the Washington and Oregon coastal fisheries than the control group (Figures 7 & 8).

Transfer Groups:

The survival rate of the 1984 brood Grays River fish transferred to Washougal was similar to the survival of the Washougal control group (Figure 9). Data for the 1985 brood are not complete. Preliminary indications are that the Grays River fish transferred to Washougal survived more poorly than the control group. The reciprocal transfer (Washougal to Grays River) survived at a similar rate as the Grays River control.

Size at Release:

Size at release of control groups varied among broods at both Grays River and Washougal hatcheries. At Grays River, the 1984 brood control group was released at 62 fpp (7.3 g) whereas the 1985 brood was released at 95 fpp (4.8 g). Survival rates are not directly comparable, but it appears that fish released at the larger size survived at a much higher rate (Figure 5). This relationship can be elucidated by comparing combined survival data of all Grays River broods by size at release in June. Survival rates of fish released a size larger than 80 fpp (5.6 g) are about 300% greater than for fish released smaller than 80 fpp (Figure 10a). Similarly, survival rates of fish larger than 80 fpp release in June from Washougal Hatchery are about 250%

higher than fish released smaller than 80 fpp (Figure 10b).

Survival rates of fall chinook released from WDF Columbia River hatcheries have generally increased since the 1983 brood. In association with the increased survival rates have been increases in size at release (Figure 11). Preliminary indications are that survival rates of 1985 brood fish, released at several hatcheries at sizes below 90 fpp (5.0 g), are lower than survival rates of fish released at larger sizes.

DISCUSSION

A study was done to assess rearing and release strategies that were alternatives to the normal rearing program at WDF Columbia River hatcheries. Increased survivals were found in June release fish exceeding 80 fpp. In fact, an apparent relationship exists between the increased size at release and increased survival within spring (including June) releases. A weaker relationship exists between release size and increased survival within fall releases. Survival rates decreased with increasingly later fall releases, even though these later released fish were larger. However, these later released fall groups still had equal or higher survival rates than fish released in June at sizes smaller than 80 fpp.

Contribution to Washington coastal fisheries is an important consideration particularly with increased emphasis placed on providing increased recreational opportunity (WDF, 1989). Contribution to the Washington coastal fisheries (percent of total survival) of chinook released in June or September (Washougal Hatchery) was lower than for groups released in October and November. However, because survival rates of the June/September groups were higher, the realized contribution of these groups (numbers or pounds of fish) to the Washington fisheries may be greater. At Grays River, increased survival and contribution rates to the Washington fisheries from the fall releases would indicate that this program would be cost effective. More rigorous analysis will be done in the future to answer these questions.

ACKNOWLEDGEMENTS

We wish to thank the National Marine Fisheries Service for providing funding for this study through the Columbia River Fisheries Development Program. We would also like to acknowledge the hatchery crews at Washougal, Grays River, and Eloomin hatcheries for their support and commitment to collecting data.

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Table 1. Pertinent release data for alternative release strategy groups by hatchery and brood.

| <u>HATCHERY</u> | <u>BROOD</u> | <u>RELEASE DATE</u> | <u>NUMBER RELEASED</u> | <u>SIZE AT RELEASE</u> | | |
|-----------------|--------------|---------------------|------------------------|------------------------|------------|----|
| | | | | <u>GRAMS</u> | <u>FPP</u> | |
| WASHOUGAL | 1982 | 8/31/83 | 101206 | 16.2 | 28 | |
| | | 10/11/83 | 100572 | 19.7 | 23 | |
| | | 11/02/83 | 100264 | 20.6 | 22 | |
| | 1983 | 6/28/84 | 101594 | 6.2 | 73 | |
| | | 8/30/84 | 100892 | 12.6 | 36 | |
| | | 9/28/84 | 101498 | 18.9 | 24 | |
| | | 10/31/84 | 101223 | 24.7 | 18 | |
| | (Transfer) | 1984 | 6/16/85 | 104802 | 4.8 | 94 |
| | | | 6/16/85 | 79590 | 4.8 | 94 |
| | | | 8/30/85 | 102750 | 13.8 | 33 |
| | | | 10/04/85 | 75698 | 18.2 | 25 |
| | | | 10/31/85 | 101872 | 23.3 | 20 |
| | (Transfer) | 1985 | 6/14/86 | 214371 | 5.1 | 89 |
| | | | 6/14/86 | 75738 | 5.1 | 89 |
| | | | 9/02/86 | 98572 | 13.4 | 34 |
| | | | 9/29/86 | 93054 | 18.2 | 25 |
| 10/30/86 | | | 94109 | 21.6 | 21 | |
| GRAYS RIVER | 1984 | 6/24/85 | 104458 | 7.3 | 62 | |
| | | 10/03/85 | 101146 | 20.6 | 22 | |
| | 1985 | 4/23/86 | 95631 | 13.4 | 34 | |
| | | 5/28/86 | 100509 | 4.7 | 96 | |
| | | 9/15/86 | 98529 | 18.9 | 24 | |
| | ELOKOMIN | 1985 | 4/25/86 | 97718 | 12.1 | 38 |
| 6/04/86 | | | 100438 | 5.9 | 76 | |

WDF, SPRING CREEK, & ODFW HATCHERIES COMPARISON OF SIZE AT RELEASE

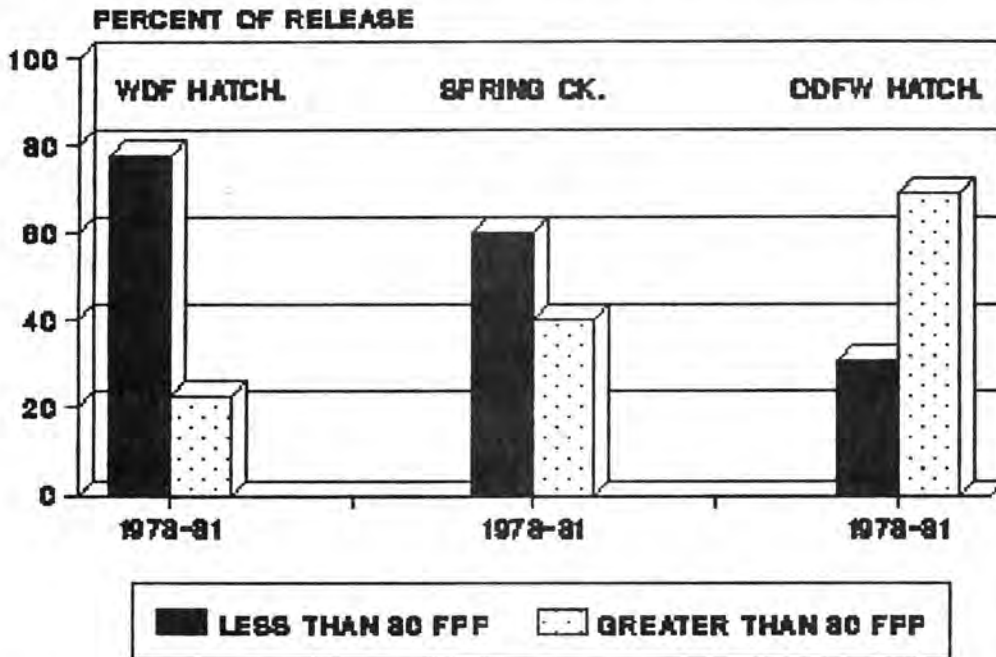


Figure 1. A comparison of size at release for chinook salmon released from Columbia river hatcheries by agency.

WDF, SPRING CK AND ODFW HATCHERIES MONTH OF RELEASE 1978-1981 BROODS

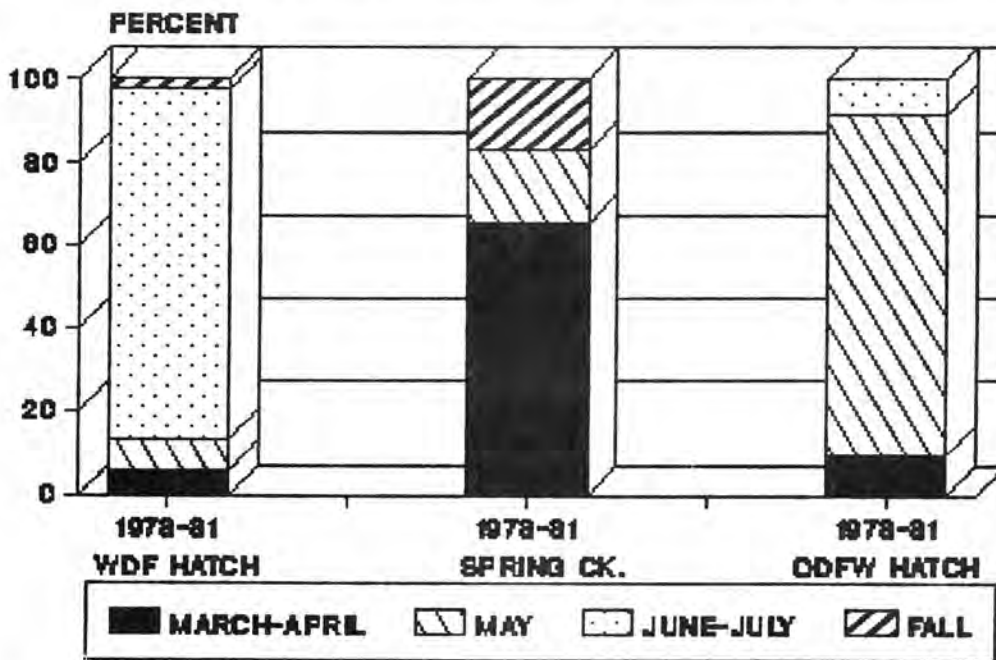


Figure 2. A comparison of release times for Columbia river fall chinook by agency.

WASHOUGAL FALL CHINOOK % SURVIVAL BY BROOD AND RELEASE MONTH

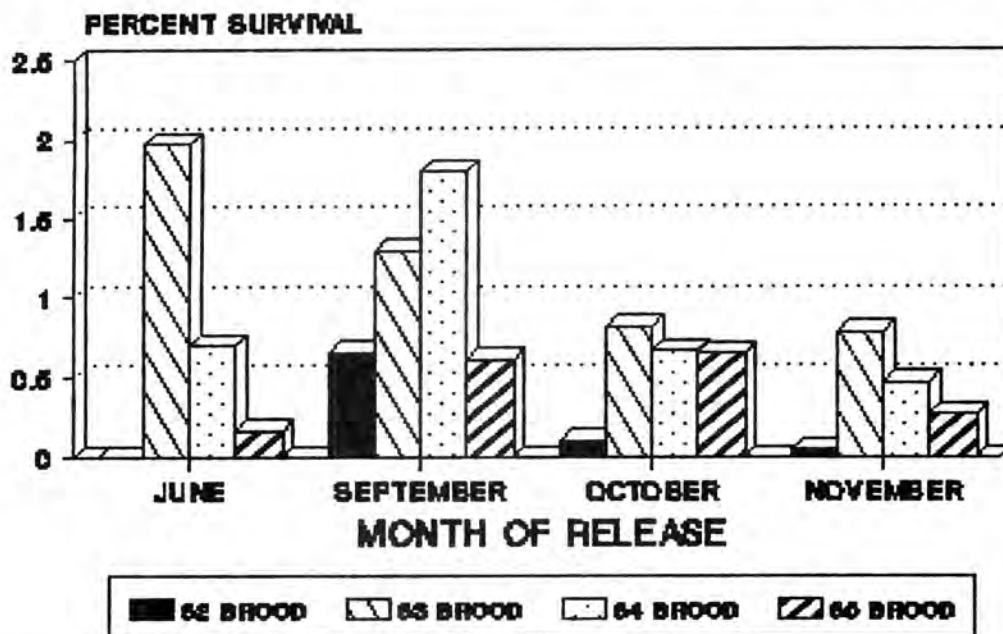


Figure 3. Survival rates by brood and release month at Washougal hatchery.

WASHOUGAL FALL CHINOOK % CONTRIB. TO OCEAN FISHERIES

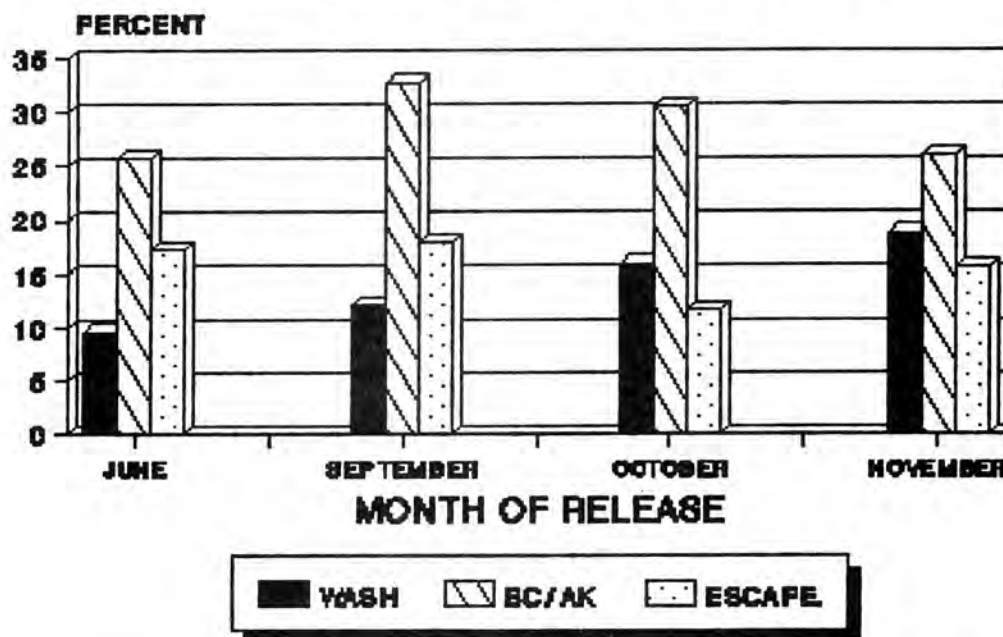


Figure 4. Contribution rates to the Washington coastal, B.C and Alaska fisheries, and escapement rates of the various release groups. Combined broods.

GRAYS RIVER CHINOOK % SURVIVAL BY BROOD AND RELEASE MONTH

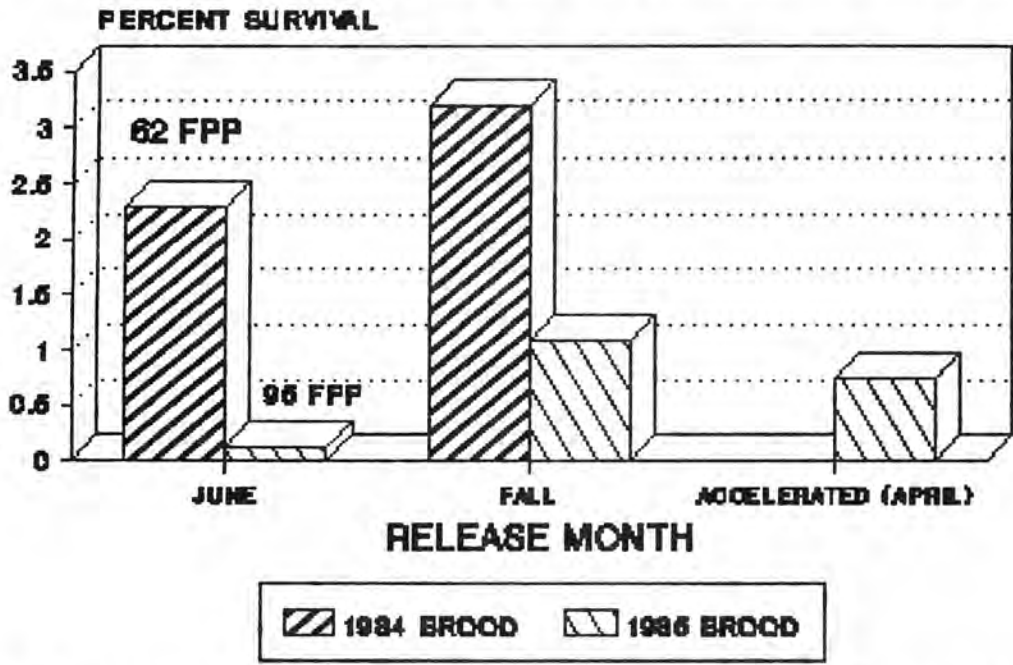


Figure 5. Survival rates of 1984 and 1985 brood release groups at Grays river hatchery.

GRAYS RIVER FALL CHINOOK % CONTRIBUTION TO OCEAN FISHERIES

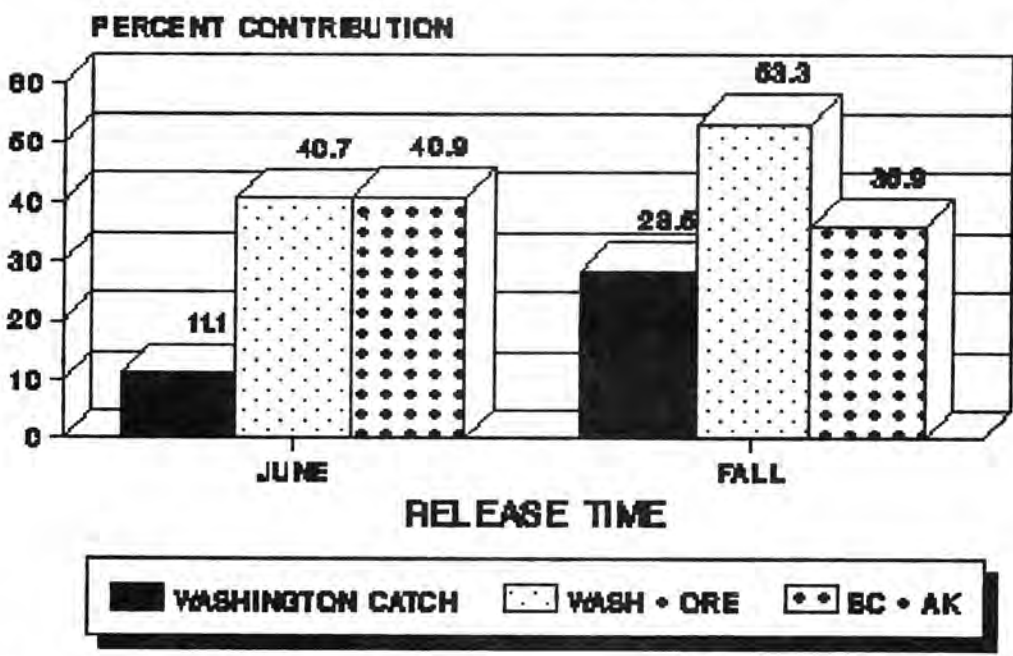


Figure 6. Contribution to the Washington, Washington and Oregon combined, and BC and Alaska fisheries from 1984 brood Grays river chinook.

SURVIVAL RATES OF ELOKOMIN FALLS 1985 BROOD

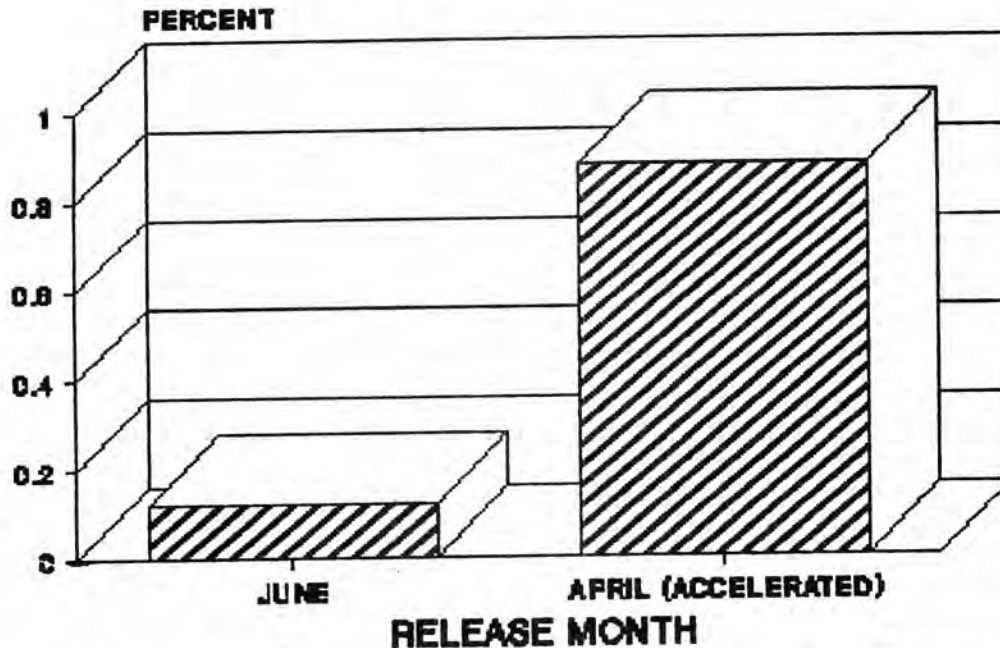


Figure 7. Survival rates of the 1985 brood release groups from Elokomín hatchery. Data is not complete.

CONTRIBUTION OF ELOKOMIN FALLS 1985 BROOD

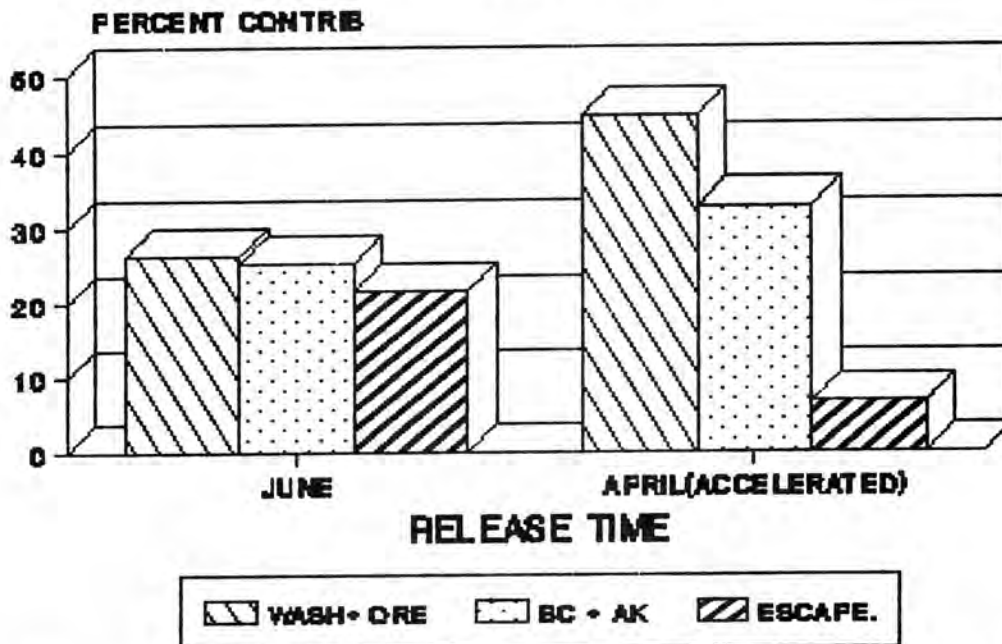


Figure 8. Contribution to the Washington, Washington and Oregon combined, and BC Alaska fisheries from 1985 brood Elokomín hatchery chinook. Data is incomplete.

WASHOUGAL AND GRAYS COMPARISON OF TRANSFER GROUPS

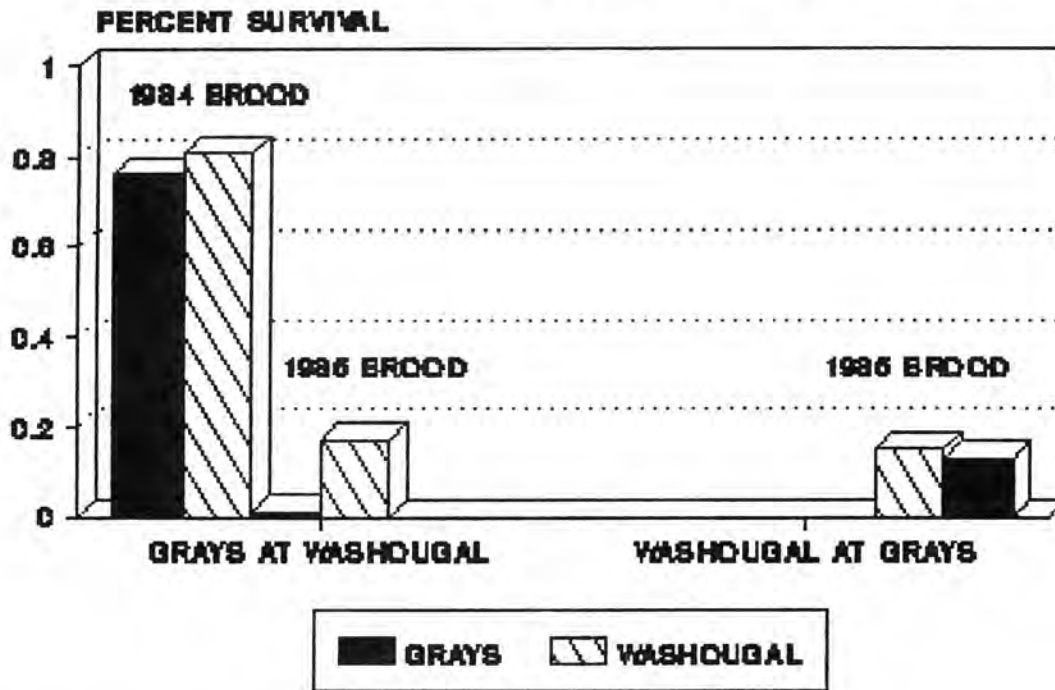
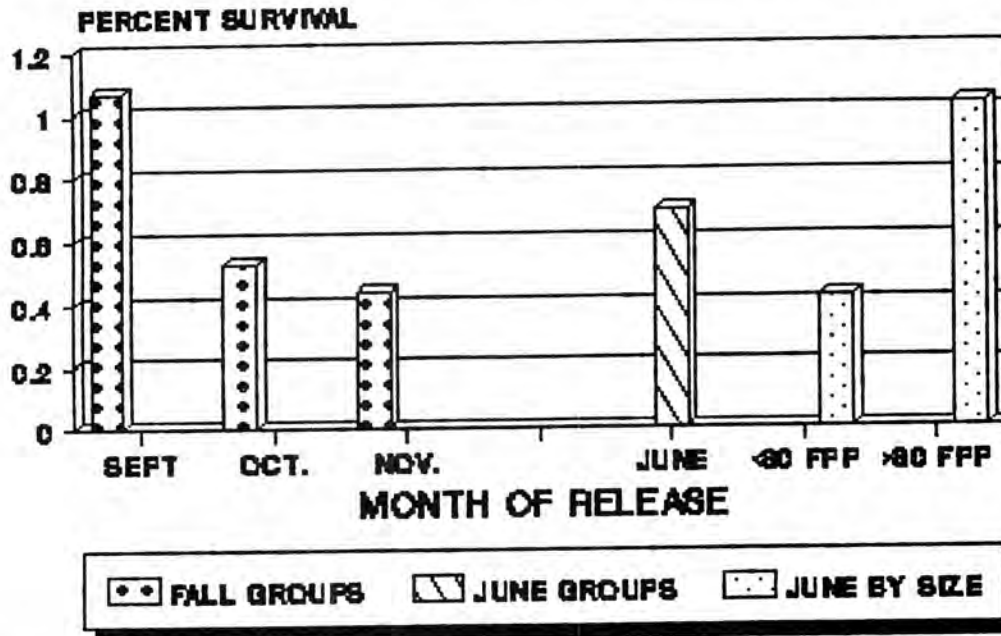


Figure 9. Survival rates of Grays river chinook (1984&1985 broods) transferred to Washougal hatchery, compared with Washougal control (June release). Also survival rates of Washougal chinook (1985 brood) transferred to Grays river, compared with Grays river control (June release).

PERCENT SURVIVAL BY RELEASE TIME AT WASHOUGAL HATCHERY



COMBINED BROODS

Figure 10a. A comparison of survival rates by release month (combined broods) and the difference in survival rates of fish released in June (by size).

GRAYS RIVER FALL CHINOOK % SURVIVAL BY RELEASE TIME AND SIZE

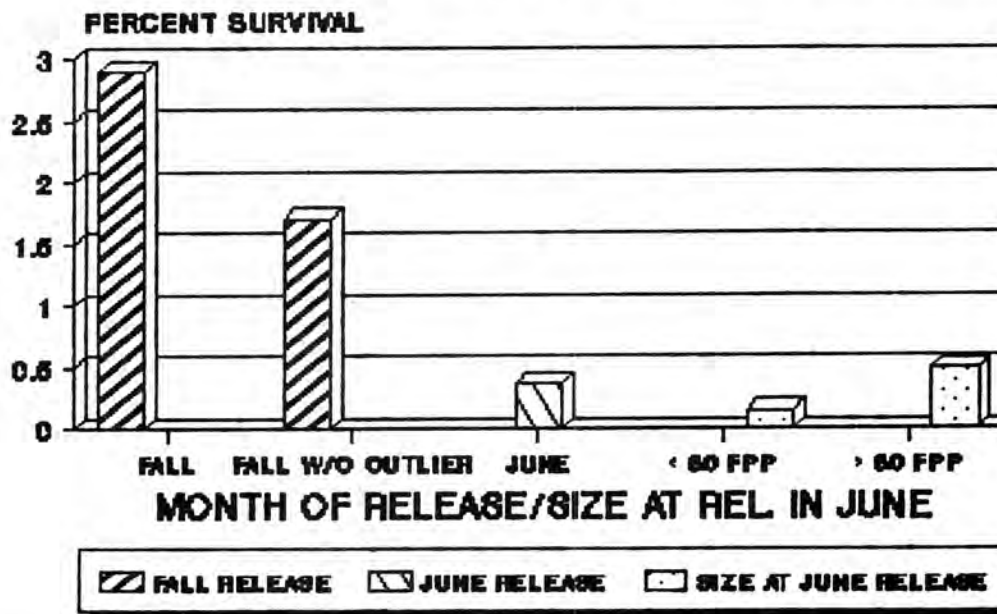


Figure 10b. A comparison of survival rates by release month (combined broods) and the difference in survival rates of fish released in June (by size). Outlier referred to in Fall release category is single tag group (8.0% surv.).

SIZE AT RELEASE VS SURVIVAL OF WDF COLUMBIA RIVER FALL CHINOOK

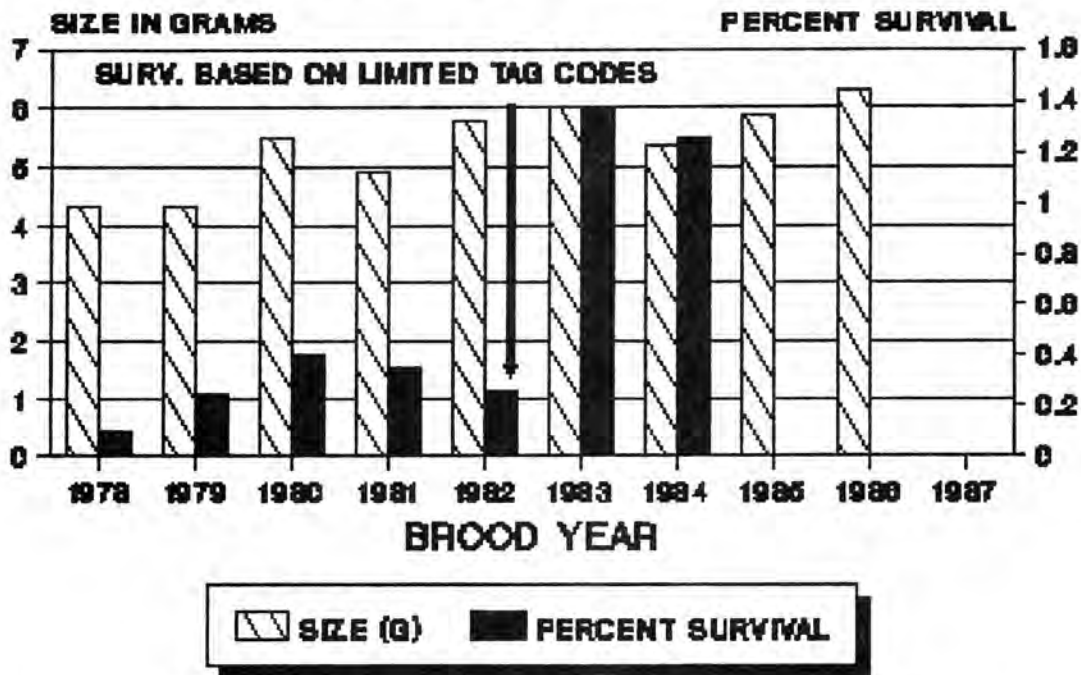


Figure 11. The relationship of size at release and survival of WDF reared fall chinook released from Columbia river hatcheries. Solid bars indicate survival rates (Right hand axis). 1982 brood had limited number of tag codes on which to calculate survival rates.

THE EFFECTS ON SURVIVAL AND HOMING FROM TRUCKING HATCHERY
YEARLING COHO SALMON TO RELEASE SITES

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ABSTRACT

We found that relative survival to adult (survival of treatment/survival of control) of hatchery coho salmon *Oncorhynchus kisutch* yearlings that were trucked and immediately released at three different locations within a Oregon coastal river basin varied from 43% to 108% over a 4-year study. Fish transported prior to release averaged 76%, 83%, and 84% of the survival of fish released at the rearing hatchery without trucking. Groups that were acclimated for 6 weeks prior to release consistently survived at a higher rate than groups released immediately after transportation. We also found that

almost all adults that returned from a release site 23 km upstream from the rearing hatchery returned to the rearing hatchery, whereas only 7% to 26% of the adults that returned from fish transported to a tributary 11 km below the hatchery returned to the rearing hatchery. Return to the rearing hatchery of fish transported to another river basin on the Oregon coast was less than 0.01%

(This study has been accepted for publication in the North American Journal of Fisheries Management).

PYRAMID LAKE'S TROUT PRODUCTION PROGRAM

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Abstract

The hatchery systems of Pyramid Lake Fisheries' restoration program are presented. These systems are:

- 1) Recycle hatcheries
- 2) Pumped lake water rearing systems
- 3) An egg collection station

The role, design, and operation of each production system is presented.

Recycle Hatchery

Introduction:

The concept of water reuse in a hatchery environment has been pursued since the 1960's. The attractive feature of hatchery water reuse was the perceived ability to grow large quantities of fish with a minimal water supply, while tailoring the rearing environment to optimize fish production (Figure 1). However, few successful recycle hatcheries have been constructed. Factors limiting adoption of recycle technology have been: inadequate system design, high cost of construction, and high cost of system operation. For a recycle hatchery to be successful, the following consideration must be met; (1) adequate flow of water, (2) efficient removal of waste products, (3) disease control, (4) temperature control, and (5) reliable backup systems. Due to a very limited water supply, and a need to produce a large volume of fish, the Pyramid Lake Fisheries program has developed a successful hatchery design which incorporates the needs stated above.

System Design and Component Function:

The strategy of Pyramid Lake's recycle hatchery system is to achieve a large volume of water flow from a very limited water supply. The flow of water in Pyramid Lake's hatchery system is as follows:

- 1) Water flows from an elevated storage tank to rearing containers.
- 2) Water flows from rearing containers to biological filters.
- 3) Water flows from biological filters to a pump gallery.
- 4) Water is pumped from the pump gallery back to the storage tank.

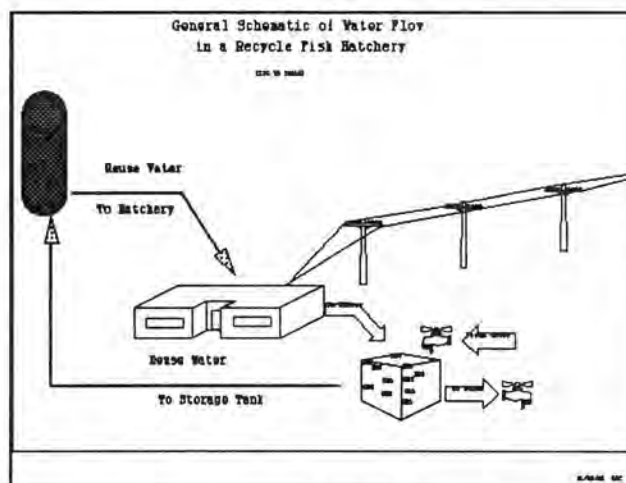


Figure 1

The components comprising the system and the functions they serve are as follows:

- 1) Wells- Ground water wells provide a continuous supply of fresh water to the system. A minimum of two wells is required.
- 2) Cooling Tower- The evaporate cooling tower lowers the temperature of the ground water to a suitable rearing temperature. The cooling tower consists of motor driven fans that move a high volume of air over finely dispersed water.

- 3) **Storage tank-** The storage tank has been found to be a key component to the successful operation of the hatchery. It provides four key functions by increasing the volume of water in the system. These are:
- A) It provides a reserve capacity of water for cleaning rearing tanks and bio-filters.
 - B) It provides head pressure which increases the velocity of water delivered to the rearing containers.
 - C) Its increased volume helps reduce swings in temperature and water quality.
 - D) Its provides a degree of emergency storage in the event of recycle pump failure.
- 4) **Rearing containers-** All the rearing containers in the system are circular tanks. The tanks provide a large surface area to volume interface which facilitates gas exchange, and they are relatively self-cleaning.
- 5) **All outdoor rearing containers** are shaded and fenced to prevent bird predation, algal growth, and solar heat gain.
- 6) **Biological filters-** The biological filters remove ammonia and nitrite by microbial action. Clinoptilolite, a zeolite material is used as a biological filter media.
- 7) **Pump gallery-** The pump gallery serves as a transfer location between water flowing from the bio-filters and returning from the storage tank. The gallery houses five vertical turbine pumps to transfer the water.
- 8) **Pump controller-** The pump controller is a digital electronic device which monitors the water level in the storage tank and the pump gallery. The controller determines how many turbine

pumps must run to maintain a balanced flow.

- 9) **Telephone alarm system-** The alarm system monitors up to 8 locations in the hatchery and will dial up to 8 telephone locations programmed in the dialer's memory when an alarm condition exists.
- 10) **Emergency generators-** Two 100 kilowatt generators are present to operate the system when a power failure exists.
- 11) **Emergency aeration-** An air blower designed to deliver an adequate air volume to rearing containers is present should water delivery systems fail.

Summary:

The incorporation of the adequate mechanical, biological, and backup systems has resulted in the successful operation of a trout recycle hatchery. A water reuse rate exceeding 90% has been achieved while maintaining total ammonia levels in the 0.1 mg per liter range.

Lake Water Rearing Systems

The lake water rearing systems consist of a pump station and onshore rearing containers. The purpose of this facility is to provide an acclimation environment where fish reared in fresh water hatcheries can become conditioned to the saline and alkaline environment of Pyramid Lake. This rearing site also provides a location to rear fish to a larger size prior to release. The need to incorporate this rearing step was based upon: (1) observations of excessive bird predation of fish released directly from fresh water facilities to the lake, and (2) data which indicated that fish size was an important factor in survival following release.

Design changes made to the lake water rearing area in recent years have been the installation of submersible pumps to replace onshore centrifugal pumps. The reason for converting to submersible pumps was to accommodate the extreme fluctuations (+/-20 feet) in the level of

Pyramid Lake. A new intake, pump station, pipeline, and rearing space was added to the lake rearing system in 1989.

Brood Collection Station

The effluent stream from the onshore rearing station had the unexpected result of attracting a spawning run. Cutthroat trout are attracted to the odor of the juvenile fish held in the onshore rearing containers as well as the temperature differential of

the effluent stream. The difference in temperature exists because the intake for the lake rearing system is below the lake's thermocline. During the late spring spawning months, there is a 5°C difference between the lake's surface temperature and that of the effluent stream. Up to 18,000 adult fish have returned to the 4 cfs effluent channel. The occurrence of this spawning run has allowed the collection of eggs from adults which have survived the harsh environment of Pyramid Lake.

NIMBUS SALMON AND STEELHEAD HATCHERY

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Rancho Cordova, CA 95670

WHY A HATCHERY

Nimbus Hatchery was constructed in 1955 by the U.S. Bureau of Reclamation to replace the spawning area lost by the construction of Folsom and Natoma dams on the American River, a tributary of the Sacramento River.

Nimbus Hatchery is operated by the California Department of Fish & Game. Funds to operate and maintain the hatchery come from the U.S. Bureau of Reclamation Central Valley Project.

NIMBUS HATCHERY PRODUCTION

Nimbus Hatchery produces 4,000,000 Chinook Smolts (60/lb or larger), 5-10,000,000 swim-up Fry and 500,000 Yearling Steelhead annually.

All smolt and yearling-size fish are transported to the Vallejo area and released into San Pablo Bay.

Chinook and Steelhead swim-up fry are released into the American River at the bottom of the fish ladder or into the Sacramento River below the mouth of the American River.

Transporting the Chinook Smolts and Yearling Steelhead around the Delta to San Pablo Bay has increased the survival from .3% to a high of 3.5%. 70% of all Chinook Salmon caught in the ocean off California come from the Sacramento River system. 40-50% of the ocean catch are from the American River.

Because of its location (Metropolitan Sacramento), Nimbus Hatchery has 350,000 to 500,000 visitors a year making it one of the most visited hatcheries in the United States. If you are in California and the Sacramento area, stop and say "Hello".

THE INSTALLATION

General Description

Nimbus Salmon and Steelhead Hatchery is located approximately 20 miles East of Sacramento on the American River directly below Nimbus Dam which, in turn, is seven miles below Folsom Dam.

Hatchery Building

The Hatchery building is 100 feet by 80 feet in size and is constructed of corrugated aluminum siding and roofing on a steel framework, as are all of the buildings. This building contains 136 aluminum hatching troughs, each 16 feet long by 16 1/8 inches wide by 7 1/2 inches deep, mounted in pairs on steel supports with storage shelves for hatchery equipment under the troughs.

Hatching baskets having a capacity of 360 ounces are used for uneyed eggs. Eyed eggs are held on standard aluminum screen frame trays with a 14 by 18 per inch mesh aluminum screen on the bottom. A 1/4 inch plastic spline was added to the perimeter of the trays to provide additional spacing between the trays for eggs. The trays are stacked nine deep, with the top tray used as a cover. Two specially designed aluminum C clamps hold the stacked trays together. Six stacks of trays are used in each trough. Each stack has a capacity of 320 ounces of eggs (liquid measure) making the capacity of each trough 1,920 ounces of eggs, or approximately 163,200 eggs based on the average size of American River king salmon eggs.

Office and Shop Building

The office and break-room are combined in one building 40 feet by 40 feet in size. The break-room occupies one-half of the building. The employee's locker room and

shower, as well as public restrooms, are in this building.

Food Processing and Storage Building

The building is 100 feet by 40 feet in size and contains the machinery for processing and storage of fish food, boot and storage area and the domestic water system. The processing building also contains the domestic water system consisting of pumps and a pressure tank with a chlorine injector.

Pond System

The pond system consists of twelve gravel-bottomed raceway-type ponds 12 feet wide by 100 feet long used principally for the rearing of steelhead. These ponds may also be used for rearing young salmon. There are four nursery ponds lined with concrete 8 feet wide, 170 feet long and 5 feet deep. These are used primarily for rearing young salmon and steelhead after they leave the hatchery building.

There are two concrete-lined adult salmon holding ponds, 46 feet wide, 157 feet long and 6 feet deep. The water enters through submerged orifices at the upper ends of the ponds. The upwelling of water from this type of entrance reduces the jumping activities of the salmon.

There is one steelhead and salmon rearing pond 68 feet wide, 201 feet long and 6 feet deep. This pond has a concrete bottom and sides. Water for this pond comes from the tail channel at the end of the nursery ponds.

Fish Ladder

The fish ladder is 502 feet in length with 30 pools, each 9 feet wide by 16 feet long and 5 feet deep. The salmon blocked by the fish rack enter the fish ladder and surmount the 20 feet of elevation differential between the river and the holding ponds.

Fish Rack And Hoist

The fish rack is 306 feet long. There are 10 concrete piers permanently imbedded in the river. The tops of the piers are 10 feet above the river bed. Nine rack support

frames, with walkways, are placed on these piers. Twenty pipe rack frames, holding the 3/4 inch galvanized pipe pickets, are then placed upon the rack support frames. A steel wire fabric mat 7 feet wide is in place 6 to 12 inches below the surface of the river bed. The pickets are driven through the river bed gravel below the fabric mat. This prevents salmon from digging under the racks.

An electric high line hoist of 5,000 pounds capacity is provided to place or remove the racks

and rack support frames. There is a 100-foot tower on the south side of the river, to which the load cable is attached. This cable extends 543 feet to the north side of the river where it is attached to a concrete block imbedded in the bluff.

Water System

Water for the installation is obtained from Nimbus Reservoir. A 42-inch concrete pipe line conveys the water from the south abutment of Nimbus Dam to a terminal structure. This pipe is capable of carrying 60 c.f.s. Thirty c.f.s is the requirement for the present plant.

During the planning of Nimbus Hatchery the conduit capable of carrying 60 c.f.s was requested by the department of Fish and Game for a possible future trout hatchery below Nimbus Hatchery. The cost of increasing the size of the pipeline from 30 c.f.s to 60 c.f.s capacity was borne by the Wildlife Conservation Board.

Nimbus Reservoir fluctuates as much as 10 feet in surface elevation, reducing or increasing the head of water at the hatchery inlet; an electrically operated automatic gate was installed in the inlet of the terminal structure to compensate for this fluctuation and provide an even volume of flow in the supply line.

Manually operated outlet gates in the terminal structure control the water flow to the pond system, hatchery and domestic water system.

A 33-inch concrete pipe carries water to the rearing pond control structure, where water may be diverted to these ponds. From the rearing pond control structure the 33-inch pipe continues to the nursery ponds. From

here the water is conveyed in an open conduit to the salmon holding ponds, steelhead rearing ponds and down the fish ladder into the river.

A 24-inch concrete pipe conducts water to two 16-inch concrete-lined steel pipes that supply the hatchery.

A 8-inch concrete-lined steel pipe conducts water from the terminal structure to pumps in the processing building. The water is chlorinated, pumped into a pressure tank, then under 70 pounds pressure distributed to all the buildings, fire hydrants and outside faucets.

Personnel Staffing

- 1 - Fish Hatchery Manager II = 12 months
- 1 - Fish Hatchery Manager I = 12 months
- 2 - Fish Culturists = 12 months
- 1 - Fish Culturist = 7 months
- 3 - Fish & Wildlife Assts = 8 months
- 1 - Fish & Wildlife Asst = 7 months
- 2 - Fish & Wildlife Assts = 12 months

Water Temperature Control

Water temperature control at Nimbus Hatchery is accomplished by removing and installing shutters in three water outlet bays in Folsom Dam. The shutters are in

nine sections numbered from bottom to top (1-9). Although there are nine shutters, the bottom seven are bolted together and must be raised or lowered as one unit. Under normal operations all nine shutters are installed in January when water temperature in the hatchery reach 45 F. With the nine shutters in place, water is now being drafted off the top of Folsom Lake. When water temperatures reach 60 F, normally in the month of June, shutters 9 & 8 are removed. This will usually stabilize the water temperature. Some time during the month of October when water temperatures at Nimbus Hatchery reach 61-62 F, shutters 1-7 are removed. The shutters are removed and installed by USBR Folsom Field Division.

Fish Trapping

The weir will be installed by the USBR during the last two weeks of September. The holding pond will be opened the end of October or when water temperatures in the hatchery reach 60 or lower. Shutters 1 through 7 in Folsom Dam will be raised prior to opening the adult holding pond.

The weir will remain in place until enough salmon eggs are taken to meet the production goals of Nimbus Hatchery and the Central Valley Project. The only time the weir would be removed prior to meeting production goal would be when water releases down the American River are expected to exceed 5,000 cfs. When flows in the American River are between 5,000-15,000 cfs, all rack panels will be removed to protect the structure.

ENNIS NATIONAL FISH HATCHERY- AN OVERVIEW

Wes Orr

U.S.F.W.S- Ennis National Fish Hatchery
180 Fish Hatchery Road
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Ennis NFH produces 25 million eyed rainbow trout eggs annually from 7 strains of rainbow trout broodstock. Eggs are available 9 months of the year.

Air spawning is used to reduce handling stress on fish and spawn takers alike. Equipment includes a small portable oxygen bottle, hose, acetylene gauge, and 3/4 inch 18 gauge needle for injecting oxygen into the fish. Working pressure is from 3 to 6 pounds depending on the size of the fish. The needle is inserted behind the pelvic fin at a slight forward angle. Try it- you'll like it!

Tests at Ennis have shown unfertilized eggs to be viable for 3 minutes in water, and 10 minutes in some saline solutions. Sperm is activated for 10 seconds in water and 15 seconds in some saline solutions. Based on several tests, we have determined 0.75% saline solution to give the best results at Ennis. We spawn directly into a 0.75% saline solution to cushion the eggs, protect them from high or low air temperatures, and to increase sperm activity time by 50%.

After fertilization, eggs are water hardened in a 75ppm Betadine solution for 30 minutes, then poured into upwelling incubators where they are treated with formalin for 15 minutes daily to control fungus. A Masterflex variable speed pump Model 7523-00 equipped with a 10 turn potentiometer is used to disperse the formalin into the incubators. Eggs are treated once a week with Betadine to control soft egg disease. A 20ppm 10 minute treatment does the job. Each incubator is also scrubbed with Betadine between batches of eggs.

Eggs are enumerated volumetrically by displacement. Regardless of the method of enumeration used, consistency is the key to accuracy.

Eggs are packed in wood-frame trays with 1/8 inch untempered peg board bottoms. They are disinfected by immersing the trays in Betadine. The trays are then drained but not rinsed before being placed in their insulated shipping cartons!

Broodfish should have adequate fat going into the spawning season. That is, 3/4 of the pyloric caeca should be covered with fat, and some fat deposition in the body cavity is all right. Grossly fat females however, do not have room for fat and developing eggs, and a great deal of pressure may be exerted on internal organs. This appears to be associated with increased mortality as the time of spawning approaches.

Broodstock at Ennis are fed a whole weeks ration by adding feed to demand feeders twice a week. Three days feed may be eaten in 4 hours and the fish are without feed for 3 days after that. This does not affect their performance as broodstock.

In a test this summer, using stabilized vitamin C in our brood diets appears to have reduced post-spawning mortality from 50% plus to about 5%! The problem is we had no control without stabilized vitamin C. More on this next year!

Reducing nitrogen supersaturation by degassing may also play a major role in reducing post spawning mortality. Obviously, reducing any external stress may reduce the incidence of fungal or bacterial invasion.

Tests were conducted in 1989 using ascorbic acid polyphosphate in the fertilization procedure and in the water hardening process. These tests in duplicate using 25, 50, 75, and 100 ppm active ingredient vitamin C showed no significant differences in survival to the eyed stage between the

controls and the test groups. Sometimes, what works at one hatchery may not work at another!

Spawning Procedures:

Air Spawning has been the preferred spawning method at Ennis for 4 years. It is used on fish as small as 2 pounds and as large as 15 pounds.

Equipment consists of a small portable oxygen bottle, a low pressure acetylene gauge, 8 feet of air hose, a stainless steel injection gun with a needle adaptor (Luer-Lock) soldered on the end of it, and an 18 gauge hypodermic needle 3/4 inch long. Excess oxygen is expelled from the fish by hand so there is no need for a vacuum system.

MS-222 anesthetized females are carried belly up to the spawning table. The "needle" man inserts the needle into the pockets at the rear base of the pelvic fin at a slight forward angle. Care is taken not to insert so close to the fin that the needle contacts the pelvic girdle. As the fish is held tail down-head up, oxygen is injected into the body cavity using 3 pounds of pressure for small fish (2-5 pounds), and up to 6 pounds of pressure for fish weighing over 10 pounds. Too much air pressure may cause internal damage or may rupture the eggs as they are expelled for the body cavity. Too little pressure will prolong the spawning process unnecessarily. After the needle is removed, it may be necessary to "shake down" the female so the few remaining eggs will flow from the upper body cavity to the vent area where they can be quickly hand stripped. Any oxygen remaining in the body cavity is removed by turning the fish tail up and manually stripping the gas.

To prevent the spread of infection, the oxygen injection needle is bathed in a solution of Betadine between injections. A fine grained sponge in a 1 pound margarine container filled with Betadine works great. The lid is snapped in place for storage. The Betadine should be replaced when it loses color.

After spawning 5 to 8 females into the pan of saline water, the milt from 3 to 4 males is added and thoroughly mixed into the

eggs. Green eggs can be left in 0.75% saline for 10 minutes with no reduction in the fertilization rate.

Fertilized eggs are set aside for a short time while another batch of females are readied for spawning. The saline is then drained off and the eggs are gently poured into a 75ppm Betadine solution where they water harden for 30 minutes.

After water hardening, the eggs are poured into upwelling incubators. No eggs are measured or counted until they are eyed. If needed, the estimated number of eggs on hand is available from historical records on the number of eggs produced per female.

All females are restripped the week following spawning. This restripping cleans out any eggs that were missed the previous week, and insures a good clean spawn without egg debris and shells the following year. At Ennis, we average 300-400 eggs per restripped female. The eyeup is usually 10-15% lower on restripped eggs.

Advantages to air spawning include: 1) with large fish, it is easier on the spawner and the fish than hand stripping; 2) it does not require the degree of experience that hand stripping does; and 3) there is less chance for rupturing eggs compared to hand stripping.

Disadvantages to air spawning are: 1) the injection needle and the compressed oxygen may present a safety hazard; 2) the air injection and apparatus increases the risk of infection; and 3) some people claim it takes longer though we have not found this to be true.

Effects of Vitamin C on Egg Fertilization:

Rainbow trout eggs are routinely fertilized in a 0.75% saline solution. Based on information received at the 1988 Northwest Fish Culture Conference, we wanted to test the effects of adding Vitamin C to the fertilization medium. On August 30, 1989, the eggs from 15 7-WvEw females were pooled and sperm was pooled from 10 7-WvEw males. The eggs were divided into two pans containing the solutions to be tested in duplicate. Each pan of eggs was fertilized immediately, water hardened, and incubated until they were eyed.

Rangens donated AaPP (Ascorbic acid polyphosphate 10.5% AI) for this test.

Results are shown as follows.

| Egg Fertilization Medium | % Eyed | % Eyed | % Eyed |
|-------------------------------------|--------|--------|--------|
| 0.75% saline | 68.0 | 72.3 | 70.1 |
| 0.75% saline + 25 ppm AI Vitamin C | 73.9 | 72.5 | 73.1 |
| 0.75% saline + 50 ppm AI Vitamin C | 70.8 | 69.2 | 70.0 |
| 0.75% saline + 75 ppm AI Vitamin C | 73.4 | 72.0 | 72.7 |
| 0.75% saline + 100 ppm AI Vitamin C | 75.5 | 71.6 | 73.5 |

The results are unclear. The variation between group A and B is more than the variation between levels of Vitamin C.

half was poured into a bucket containing 75 ppm Betadine plus 50 ppm Vitamin C. The eggs were water hardened for one hour.

Water Hardening Eggs in a Solution with Vitamin C:

On the same day, another group of 47 females was spawned and fertilized in the same manner. One-half of these eggs was water hardened for one hour in 75 ppm Betadine and the other half was water hardened in 75 ppm Betadine plus 100 ppm Vitamin C.

At Ennis, rainbow trout eggs are routinely water hardened in a solution containing 75 ppm AI of an Iodophore. Based on information received at the 1988 Northwest Fish Culture Conference, we wanted to test the effects of adding Vitamin C to the water hardening solution.

The test was repeated September 20, 1989, when 37 Erwin X Arlee females were spawned and fertilized in the same manner. They were divided into three groups and water hardened in 75 ppm Betadine, 75 ppm Betadine with 50 ppm Vitamin C, and 75 ppm Betadine with 100 ppm Vitamin C of for one hour. The results are shown below.

On September 13, 1989, a group of 26 females was spawned into a 0.75% saline solution and the eggs were fertilized with pooled milt from a similar number of males. After rinsing, approximately one-half of the eggs was poured into a bucket containing 85 ppm Betadine and the other

| Water Hardening Medium | % Eyed |
|---|--------|
| Betadine Solution | 74.91 |
| Betadine Solution plus 50 ppm (AI) Vitamin C | 76.93 |
| Betadine Solution | 87.63 |
| Betadine Solution plus 100 ppm (AI) Vitamin C | 85.67 |
| Betadine Solution | 81.85 |
| Betadine Solution plus 50 ppm (AI) Vitamin C | 82.06 |
| Betadine Solution plus 100 ppm (AI) Vitamin C | 82.52 |

Ascorbic Acid Diet Test

For many years, post-spawning mortality of rainbow trout has resulted in losses of 25 to 90% of male fish and from 2 to 25% of female fish. The loss is characterized by frayed fins, loss of external mucous as evidenced by feel, and then the appearance of circular rings of fungus which grow to cover the fish in just a few days. As the integrity of the skin is compromised by fungal invasion, internal osmotic balance is impossible and mortality can be severe. Therefore, we wanted to find out whether or not Vitamin C as a feed additive would affect post-spawning mortality.

In 1989, a diet test was conducted comparing mortality, growth, and egg production of Erwin broodstock fed 100 ppm (AI) Vitamin C versus 500 ppm (AI)

Vitamin C in their diet. The GR-6 formulas were manufactured on 4/19/89 by Murray Elevators. AaPP (Ascorbic acid polyphosphate) was donated by Rangens.

During April 1989, 5,480 two-year-old Erwin rainbow trout were equally divided in six raceways. Three raceways were fed feed containing 100 ppm Vitamin C. Prior to spawning, the feeding rate was based on a hatchery constant of 13.0 and a length increase of 0.70"/month. After spawning began, the fish were fed a reduced amount by eye but each group received the same amount each day. The test was conducted for 156 days beginning 4/24/89 and ending 9/27/89. The fish were sample counted 4/24/89, 6/26/89, and 9/27/89. Spawning began 6/26/89 and ended 9/15/89. Results of the test are shown below.

| | Feed with 100 ppm Vitamin C | Feed with 500 ppm Vitamin C |
|----------------------------|--------------------------------|--------------------------------|
| Number Start 4/24 | 2740 | 2740 |
| Average Weight lbs. 4/24 | 3.61 | 3.53 |
| Average Weight lbs. 6/26 | 4.12 | 4.04 |
| Average Weight lbs. 9/27 | 4.18 | 4.18 |
| 156 Days total mortality | 125 | 128 |
| Average % eyed | 76.33 | 75.75 |
| Average # eyed eggs/female | 3312 | 3199 |
| % Total Mortality | 4.56 | 4.67 |

There was no significant difference between the two groups in mortality, growth rate, and egg production. External appearance of both groups of fish was good throughout the test and mortality was very low. Most of the mortality was due to handling and anesthetizing during sorting and spawning. Post-spawning mortality, particularly among male fish did not occur during this test and was still not evident one month after the end of the spawning season. Average total mortality was only 4.60% compared to 30 to 90 % in prior years. Average percent eyeup was about 6.0% higher and eyed eggs per female was 10% higher than in prior years. Since there was no control group, it is not possible to determine whether the Vitamin C was responsible for these improved results. However, the stabilized form of Vitamin C, even at 100 ppm, may be more beneficial

than other forms of Vitamin C which break down rapidly in manufacturing, handling, and storage.

Formalin Treatments Made Easy and Safe

The use of formalin to control fungus growth on eggs can be labor intensive at large stations. Exposure of employees by inhalation or direct contact is a safety hazard.

A timer controlled, formalin contained metering system can save time and eliminate some of the hazards associated with formalin.

The Masterflex Pump, Model 7523-00, is a variable speed pump with a 10 turn potentiometer for making minute flow adjustments. A wide range of pump heads

and tubing sizes makes the unit very versatile. At Ennis, we selected size 16 Tygon tubing (N-06409-16) and a quick load head with a stainless steel rotor (N-07021-24).

By determining the flow through each jar, the desired volume of formalin can be calculated for a given concentration. Just set the flow rate with the potentiometer and the formalin is pumped directly from the 55 gallon barrel into the main incubation supply line through a continuous length of tubing. The pump head rollers squeeze the tubing as they turn, causing a peristaltic action that transfers the liquid. No mess, no smell!

Suggestions:

1. To prevent wearing a hole in the tubing, move the tubing in the pump head once a week to change the location of the rollers.

2. Check the timer each day to make sure it goes on and off. Sometimes things happen!

3. Calibrate the system and make a chart for your particular situation. The digital readout will not read accurately for the lowest to the highest flow rate. Your chart will correct that.

4. Make sure the pump head is turning the right direction. The pump has a forward and reverse switch.

5. Use the time you save to do something good for your hatchery!

DECADE OF SOCKEYE SALMON ENHANCEMENT IN ALASKA

Lorne E. White
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Fisheries Rehabilitation, Enhancement,
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The Alaska Department of Fish and Game (ADF&G) and the Private Non Profit (PNP) hatcheries have made some significant progress in the area of ocean ranching over the past 10 years. This ocean ranching program has been built with the purpose of enhancing and rehabilitating Alaska salmon stocks for all user groups. The area we would like to focus on for this conference is the sockeye salmon fish culture technology.

In 1976 and 1978, Alaskan voters approved significant bond issues for hatchery construction. There was a strong conviction at that time that the investment would pay off and, with oil money in the bank, it appeared to be a good investment. As sockeye salmon have always been considered to be the "money" fish in Alaska, this species was an integral part of the ocean ranching program. As a consequence of a conviction that the program would work and having an exceptional resource base to work with, today Alaska leads the world in sockeye salmon culture technology and is in the forefront of in-lake population dynamics of rearing sockeye salmon juveniles.

The background for sockeye salmon fish culture in Alaska is exceptional in that we have:

- 1.) An excellent management strategy which has allowed for large escapements and catch, permitting sufficient broodstock for enhancement.
- 2.) Excellent water quality for hatchery use and rearing environment.
- 3.) An exceptional research staff to draw upon for life history data.

- 4.) Diverse genetic gene pool of sockeye, ie. early and late stocks and multiple age groups.

Hatchery program:

Today, ADF&G and PNP hatchery programs are incubating roughly 100 million sockeye eggs between 8 facilities. The largest facility, in terms of number of eggs incubated, is the Gulkana stream side system in Interior Alaska. It incubates approximately 35 million eggs each year, while the other facilities incubate in the neighborhood of 20 million or less. Some facilities incubate less but release fish that are reared for a longer period, such as Kitoi and Main Bay. Our programs have come a long way over the past 10 years.

During the period of 1978 to 1982, we were really having some significant problems. The problem was of course infectious hematopoietic necrosis (IHN) virus. The challenge was to create a culture process that could still produce fish in an uncertain environment. We went ahead with our programs knowing that only by doing the work would we find some answers. During the first years of the program, keeping the juvenile sockeye salmon alive was a hit and miss affair. The East Creek Hatchery at Dillingham and Big Lake Hatchery near Palmer had some significant losses to their annual production. At the time, as we had an epizootic, the policy was to destroy all the sockeye salmon at the facility. It was a very depressing time for those hatchery managers.

During the winter of 1980-1981, a team of pathologists and hatchery managers set about looking for patterns or possible causal relationships. The short track

record we had showed that some facilities were still in production while others were suffering without production. There was no apparent lack of the disease in facilities that had no sockeye salmon in the water supply. While the facilities that had problems also had sockeye salmon in the water supply. The pattern of virus in the water supply and poor survival, seems now to be simplistic. However, at the time, the scientific community was looking at such things as breeding for resistance and developing vaccines. The solution we chose was prophylactic, ie. separating the virus from the fish as much as practical.

The approach we chose was to : (1) use only hatchery water that was from wells or "hanging" lakes which had no sockeye salmon; and (2) incorporate steps in our egg taking and incubation process which isolates the fish from the virus to the maximum extent possible. The second step was accomplished by spawning individual fish and treating the eggs with an iodine-like solution before the eggs were pooled and to further isolate incubators and raceways to the smallest denominator that was practical. In this manner, horizontal and vertical transmission of the disease was minimized.

Today we are incubating over 100 million sockeye eggs in eight facilities across the state, and have been doing so over the past several years. In 1987 to 1989, we lost less than 5% of our annual production to the IHN virus. This is what we call our "farm-around" the problem process. However, this achievement was only part of the program. As culturing sockeye salmon was still considered risky, we had to stock these fish out as soon as possible each year to avoid crowding and stress. Planting the fish out into lakes was the strategy. Evaluating the lake environment became the next goal to complement the hatchery stocking plan.

Lake Limnology:

Research had been carried out on Alaska lakes for over 50 years, and it was quite apparent that each lake had its individual profile for productivity. What was needed was a predictive model for sockeye salmon systems so that the appropriate numbers of fry could be stocked into the lakes

without damaging the carrying capacity of the system. By using performance data from a variety of lakes, we developed a model for lake stocking which, coupled with seasonal sampling, has helped predict the level of fry stocking each year.

Lakes limited by low numbers of young fish were classified as "recruitment-limited". These lakes could be limited by escapement or by spawning area. For example, Karluk Lake in the period of 1940 until 1985 was escapement limited and was not producing enough fry to meet the carrying capacity of the lake. Management strategy of closures on the fishery and enhancement by massive eyed egg plants has led to the success of restoring the escapement to levels not achieved since the 1930's (over 1,100,000 sockeye in 1989).

Lakes limited by the quality and quantity of the forage productivity in the rearing area are termed "rearing-limited". Forage limited systems may result from a lack of the right kind or amount of zooplankton or insects. Also, there may be a spatial or temporal barrier between the fry and the lake's food source. Environment limited systems may have an unfavorable temperature regime or have too short a growing season. An example of a rearing-limited lake would be Frazer Lake on Kodiak Island. Frazer Lake has more spawning area available for adult sockeye than can be supported by the lake rearing area. After several years of over-escapement to the system, we found that the food base of the system was damaged (Figure 1). We corrected the system by cutting back on the escapement levels into the lake and fertilizing. This year (1989) over 800,000 sockeye were caught at Frazer and 360,000 escaped into the lake. Young sockeye in the lake are in good condition and the zooplankton appears to be improving in quantity and quality.

Our investigations have linked the volume of a lake that receives transmission of light sufficient to drive photosynthesis to the base of the food web and, in turn, the sockeye salmon production. The use of this lake classification scheme has allowed us to match the appropriate enhancement strategy to the limiting feature of the lake. Some lakes we are planting with

fry, while other lakes we are fertilizing. We monitor lakes that are being enhanced and make annual adjustments to the level of stocking and fertilizing because lakes are recognized as being dynamic in their productivity. For other lakes, we may recommend no enhancement strategy at all, but may recommend a different escapement strategy to the management staff (Table 1 and Figures 2-4).

New Programs:

The Main Bay Hatchery in Price William Sound is breaking new ground in sockeye culture by incubating and rearing sockeye salmon to the yearling smolt stage. This spring, a total of 4.5 million smolts were released from this hatchery site. The program was built around dealing with small lots of fish and strict isolation of lots coupled with stringent prophylactic treatment of eggs in the egg take and incubation processes. At current ocean survivals of 20%, we are anticipating a return of 900,000 adult sockeye to this program.

The Kitoi Bay Hatchery in the Kodiak area is also breaking into some new fish culture research by incubating natural occurring underyearling sockeye salmon. Underyearling sockeye migrate to sea after only 3 to 4 months of fresh water rearing. The benefit of raising high value sockeye salmon in a short period may pay dividends in areas that are short on natural rearing space. At Kitoi, a pilot program raised 140,000 smolts to 2.5 gram size in 1988-1989. This year, we expanded the goal to a release of 4.0 million underyearlings smolts in 1990.

In closing, I'd like to say that there are excellent economic returns to be made in sockeye salmon enhancement. This year (1989) a total of 1.1 million sockeye salmon returned to Kodiak Island's Frazer Lake system. This generated 800,00 sockeye or \$7.0 million to our fishermen. This was a system which was barren to any anadromous fish until the 1950's. I think Frazer is a testimony to the potential that sockeye enhancement offers for other areas of the Pacific Coast.

Table 1. The euphotic volume (EV) units are an index one step removed from the sockeye forage base - since other factors such as flushing rates also determine forage production. It is also important to look at the zooplankton biomass.

| Lake | Surface Area | Mean Depth | EZD | EV Units | Zooplankton Biomass |
|---------|--------------|------------|------|----------|-----------------------|
| Afognak | 5.3 km | 8.6 m | 12 m | 46 | 154 mg/m ² |
| Akalura | 4.9 km | 9.9 m | 11 m | 48 | 364 mg.m ² |

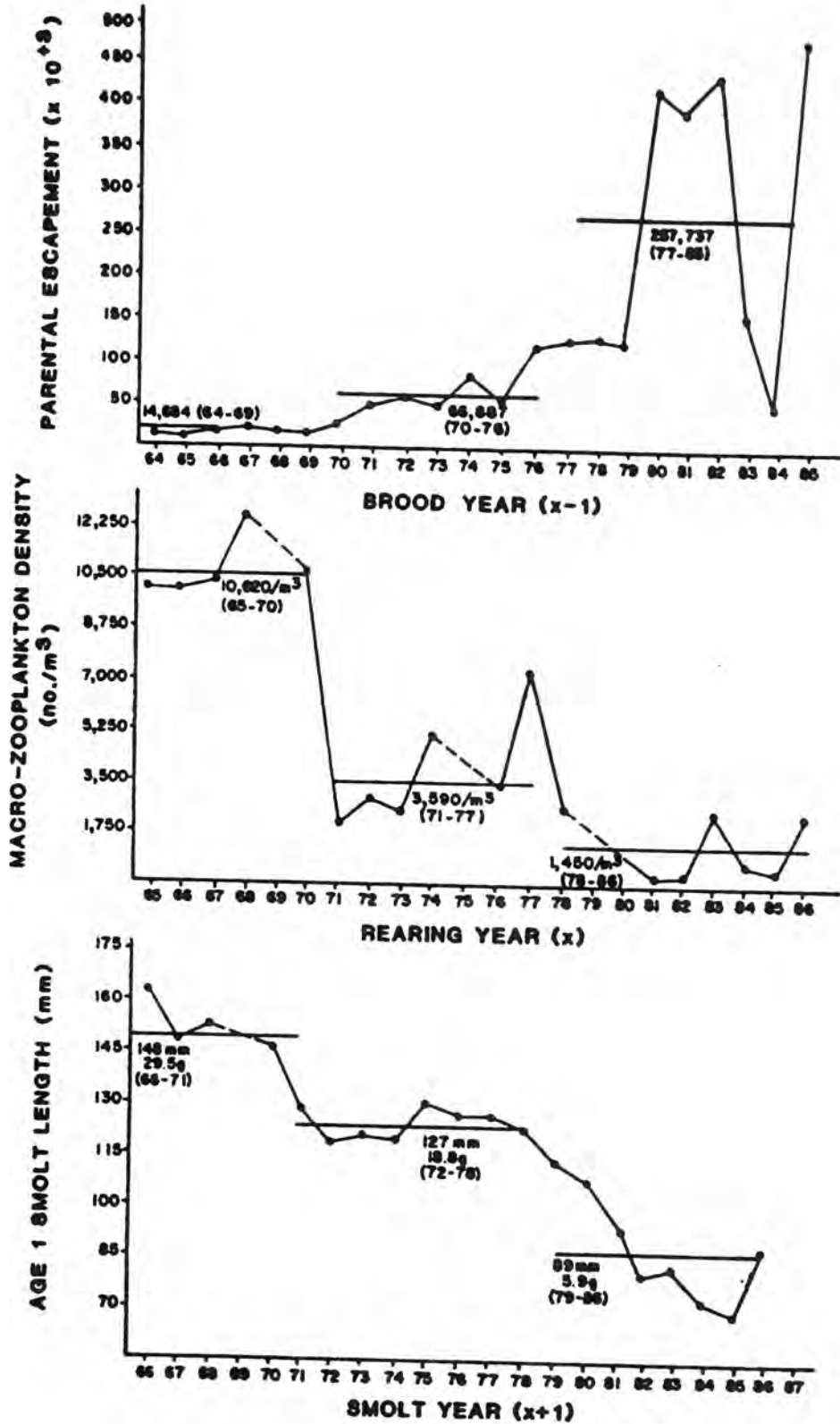


Figure 1. Frazer Lake sockeye salmon - figure shows the effect of increased numbers rearing sockeye on the forage base (zooplankton) of a lake, and the resulting reduction in smolt size.

Simplified Lake Ecosystem

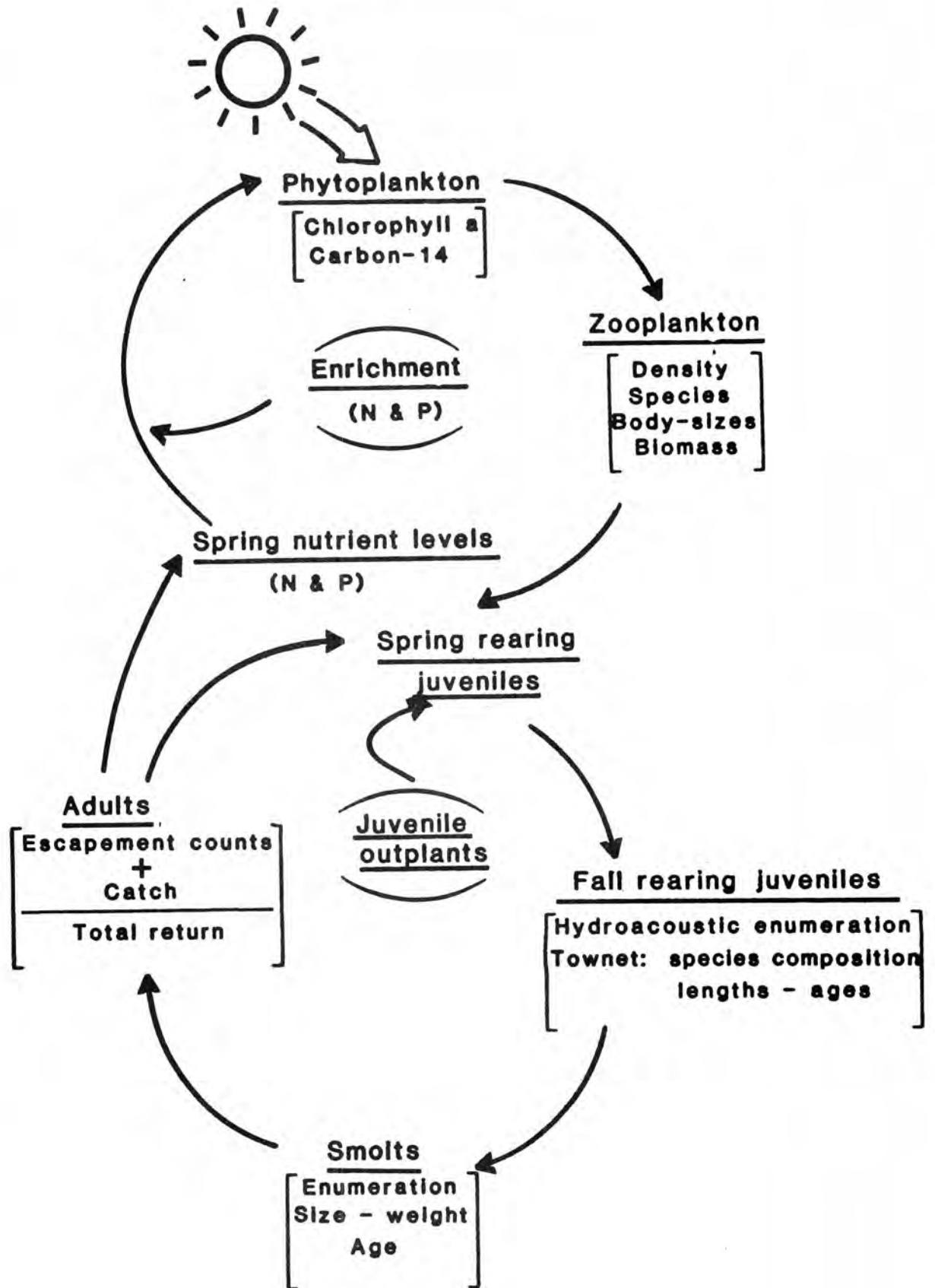
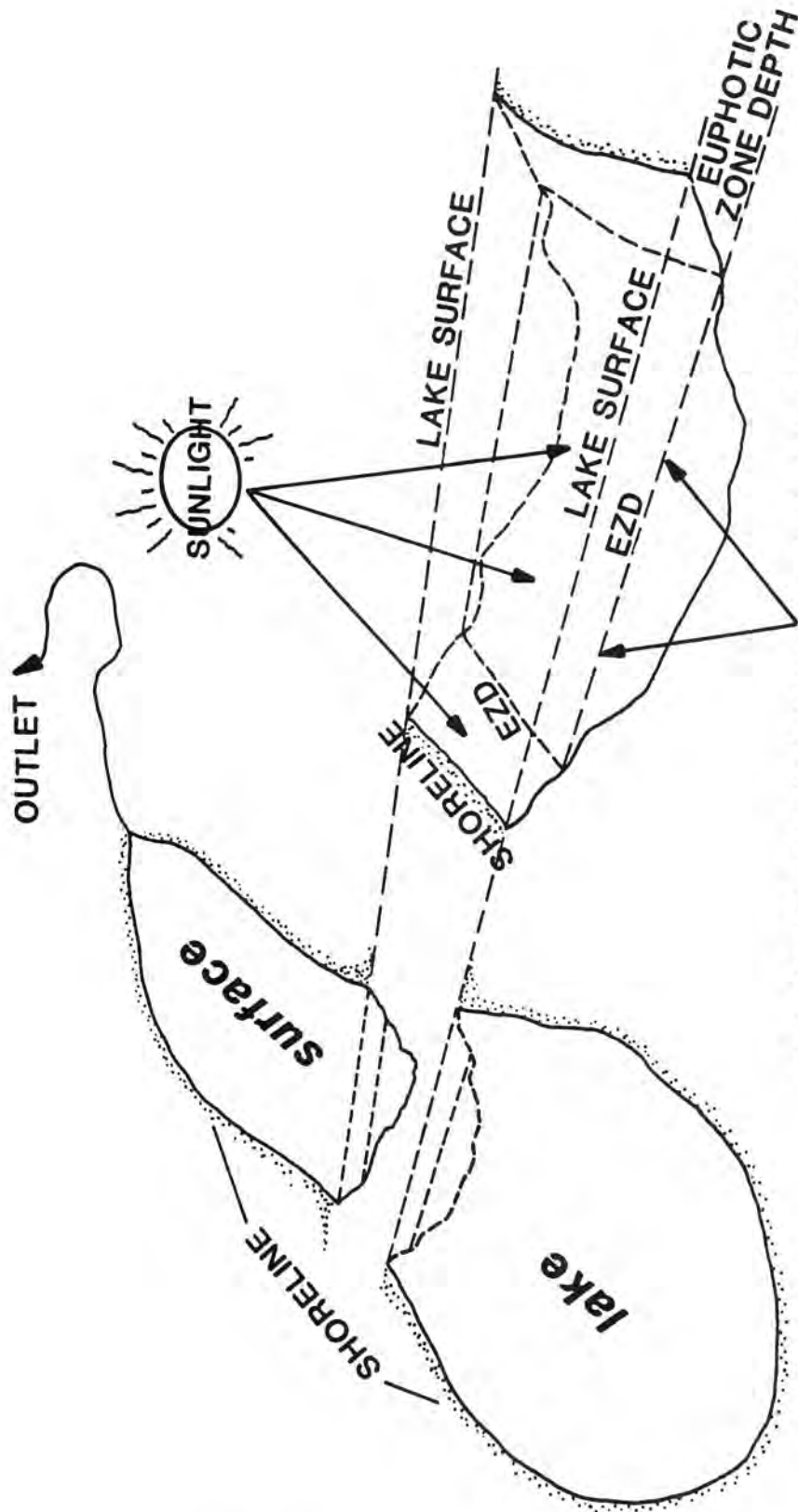


Figure 2. Evaluation procedure followed to link the lake fertility (carrying capacity) and the sockeye salmon life cycle.



DEPTH BELOW WHICH LESS THAN 1% OF THE
 SUBSURFACE PHOTOSYNTHETICALLY ACTIVE
 RADIATION PENETRATES

$$\text{EUPHOTIC VOLUME} = \text{EUPHOTIC ZONE DEPTH (EZD)} \times \text{LAKE SURFACE AREA}$$

Figure 3. The euphotic volume of a lake is linked to the carrying capacity for juvenile sockeye salmon.

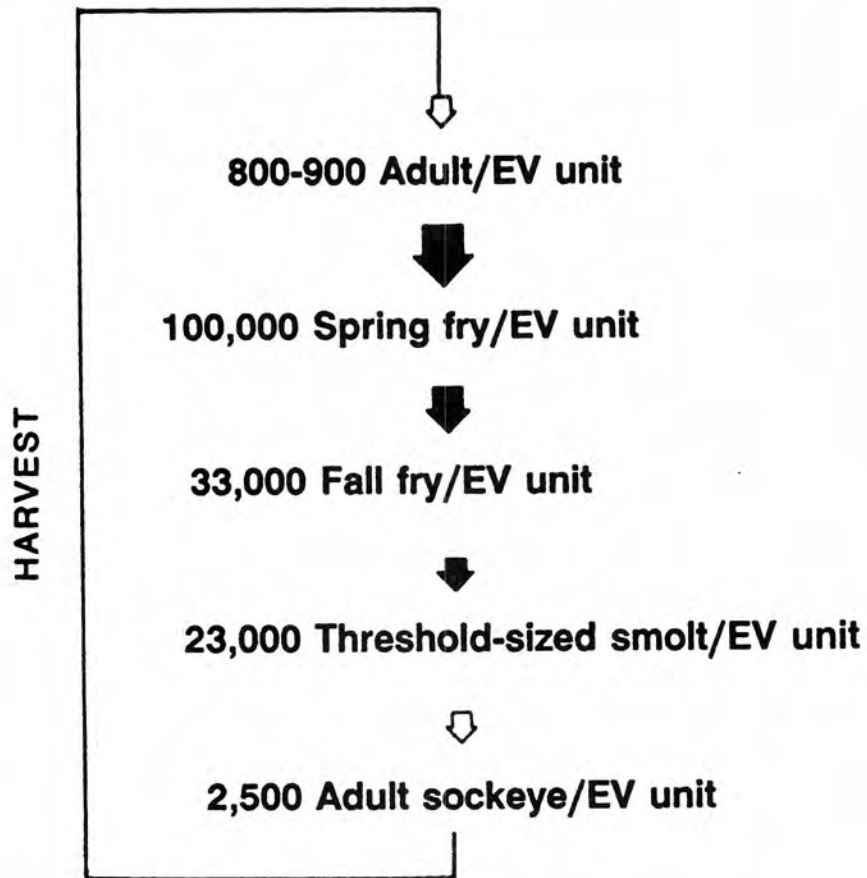


Figure 4. The sockeye salmon life cycle is linked to the lake fertility or EV (euphotic volume units) of each lake.

MUCKLESHOOT INDIAN TRIBAL FISH ENHANCEMENT FACILITIES

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The Duwamish-Green River empties into Puget Sound at Seattle. This once dynamic river system had a drainage of over 1643 square miles prior to 1900. Drawing 1 shows that major tributaries included the Black, Cedar, Green and White rivers. Between 1900 and 1916, the Cedar River was diverted into Lake Washington to flow through the government locks at Ballard and the White River was dammed at Auburn and forced to flow south to meet the Puyallup River at Sumner. Drawing 2 shows the 483 square miles of watershed left of the Duwamish. Historical discharges are shown in Plate 1.

My slide presentation takes you on a tour of the present day Green and White rivers from salt water to flood control dams and gives a brief look at tribal hatcheries of those rivers.

The first dam on the White River is Puget Sound Power and Light Company's diversion dam located at river mile (RM) 24. Water is diverted south to a hydropower project and re-enters the White River at RM 3. Five and a half miles upstream is the Army Corps of Engineers (Corps) Mud Mountain Dam, operated for flood control.

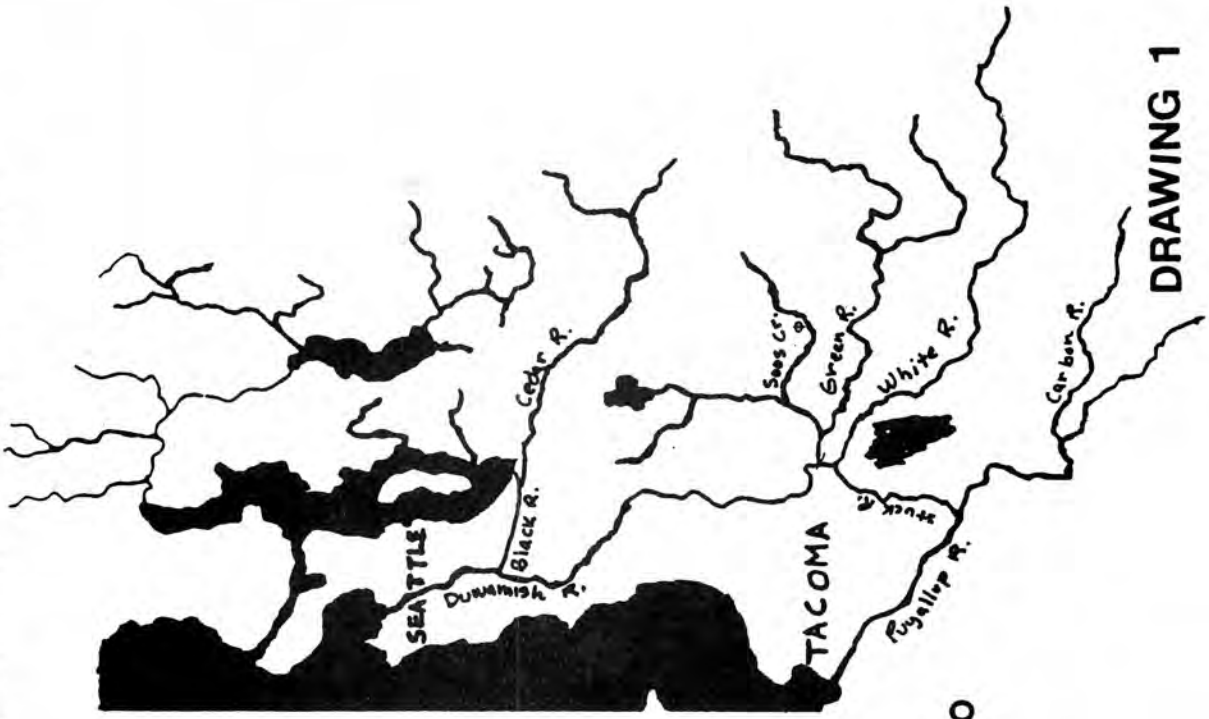
The first obstruction on the Green River is Tacoma's dam at RM 61, diverting half the river south to Tacoma for domestic and industrial uses. Three and a half miles up-stream is Howard Hanson Dam, a flood control structure operated by the Corps.

Man-made obstacles and water diversion has hampered salmon and steelhead runs for decades. The Muckleshoots and state agencies have built artificial production facilities to help mitigate for some of the losses.

Keta Creek Hatchery is located on Crisp Creek, a tributary of the Green River, entering near RM 39. The tribe built the salmon and steelhead facility between 1978 and 1982 and operate it for rehabilitation purposes. Three million fish are produced annually with the majority planted in the watershed above Howard Hanson Dam. Pumped creek water is sterilized with U.V. before flowing through packed columns for aeration. A re-use system is employed when turbidity or disease problems necessitate its use. In that loop, water also flows through clinoptilolite beds for ammonia removal.

The White River Hatchery was built in 1988 and 1988 for the tribe by Puget Sound Power and Light Company. The result of a water diversion dispute resolution, the hatchery contains state-of-the-art computer alarm/operation systems and waste management treatment. The tribe has dedicated the facility to rehabilitation of the White River spring chinook, the last viable run for springers in South Puget Sound. The 5 c.f.s. hatchery will produce approximately 350,000 zeros and yearlings annually and is scheduled to be expanded to 10 c.f.s. in 1994.

DUWAMISH WATERSHED MAP



PRIOR TO
1916

DRAWING 1

DUWAMISH WATERSHED MAP

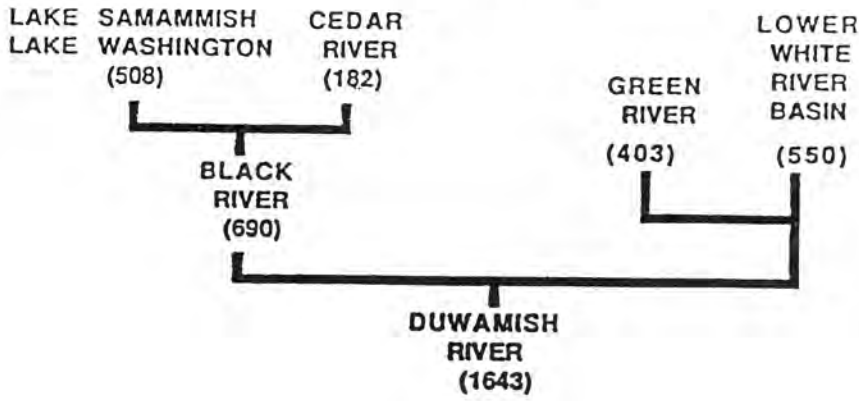


AFTER
1916

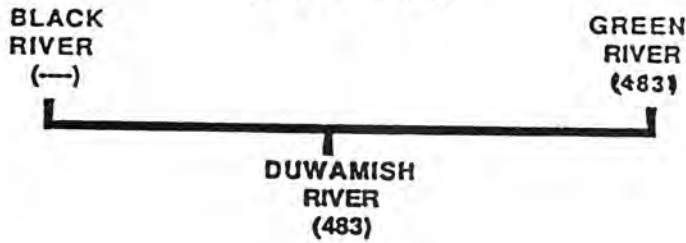
DRAWING 2

HISTORIC CHANGES IN DUWAMISH RIVER WATERSHED (sq.mi.)

HISTORIC [PRE 1900]



PRESENT [POST 1916]



HISTORICAL CHANGES IN DISCHARGE OF THE DUWAMISH RIVER 1860-1986

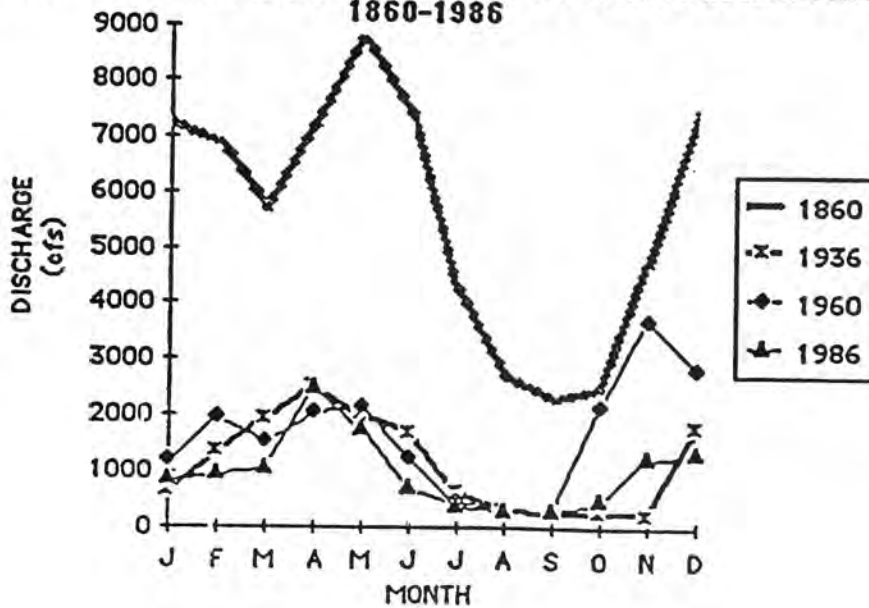


PLATE 1

CONTROLLING BIRD PREDATION ON ONE-THIRD ACRE REARING PONDS BY TOTAL COVERAGE WITH NETTING- 1989 RESULTS

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ABSTRACT

The Arcata wastewater-seawater salmonid fish culture system was invaded in 1986 by cormorants causing a significant but unquantified loss of juvenile fish on an uncovered 0.15 ha (0.33 acre) rearing unite (Winter Pond #1). To reduce loss of fish to birds, in 1987, the first of two Winter Ponds was completely enclosed in nylon netting, and the second was enclosed in 1988. In 1989, effectiveness of the netting was tested using large domestic rainbow trout (8.0 per pound). In Winter Pond #1, a lot of smaller, vibrio-vaccinated trout were used in the test. Winter Pond #1 trout were fed pellets. Smaller trout showed an 85% mortality over four months of winter rearing while larger trout only had a 13% mortality. Surviving small and large trout lots were both doubled in size during rearing. Large trout only were released in Winter Pond #2, and were not supplementally fed. Unfed trout show no growth, had a net loss in production, and suffered a 29% mortality. Netting was successful in excluding virtually all bird predators except black-crowned herons (*Nycticorax nycticorax*). Herons behavior inside Winter Pond #1 in 1988 was determined by direct observations. Heron predation in 1989 was inferred from the rate of occurrence of "beak marks" on samples of trout. Covering ponds with netting was correlated with lowered mortality rates. Mortalities in 1989 appeared more related to other factors the bird predation despite the occurrence of intense short-term predation events.

INTRODUCTION

Avian predation at the Arcata wastewater aquaculture system has been described previously along with a description of the facilities (Allen and Couch 1988). Losses at fish culture facilities to birds can be excessive (Parkhurst et al. 1987; Avault 1988). Site specific management techniques to reduce avian predation include noise, patrol (dogs, humans), lighting, visual scare devices, altering design of facilities, killing predators, and either partially or totally enclosing fish-rearing facilities. Historically, identified animal predators have been exterminated. Granting federal permits required for killing predators can be delayed by the public review and hearing process and thus are usually ineffective in emergency situations. Since the Arcata fish culture facility is in the middle of a nationally-recognized wildlife sanctuary complex, predator control and management must be sensitive to public opinion. After attempting most of the usual techniques with only limited success, we instituted a program of covering all ponds. Smaller "Summer" Ponds were netted when first constructed, but our large "Winter" (or Yearling) ponds of about 0.15 ha (0.33 acre) surface areas were not. In 1988, we netted Winter Pond #1 and in 1989 covered Winter Pond #2. Summer Ponds were also edged with 3-foot high woven wire fencing and a one-strand electrified wire (4,000 volts, low amperage) installed at a 4-inch height on planks laid immediately adjacent to the base of the fence. The woven wire fence and one-strand electrified wire were

primarily aimed at excluding otters which are abundant in the surrounding aquatic habitats.

This paper gives the results of rearing juvenile trout from January through May 1989 in the two completely covered ponds. Trout used in the study were reared in Summer Pond #1 from April 1988 through January 1989, and results of this rearing are also reported here. An assessment of the effectiveness of covering ponds is also presented as well as observation on behavior of black crowned night herons.

MATERIALS AND METHODS

Ponds:

Ponds were enclosed with black plastic netting of 10 cm. square mesh. Both ponds were enclosed as previously reported (Allen and Crouch op. cit.). Both Winter Ponds contained an array of artificial reefs made of brush-bundle platforms, car tires in various configurations, and mounds of large drain pipe surrounded with broken concrete pieces. Vertical pond banks were made either of broken concrete pieces of car tires filled with dirt and rubble. Both artificial reefs and pond banks provide habitat for fish food organisms. Water from Winter Pond #1 was pumped into a smolt trap and then discharged into the northwest corner of Winter Pond #2. Water returned to Winter Pond #1 by a drain connecting the ponds mid-way along their common bank. *Nannochloris sp.* dominated pond waters ("green water"). *Enteromorpha* was planted into Summer Ponds throughout the summer but no viable population of the marine

plant was established which, when dominate, produces a relatively "clear" pond water.

Only a single Summer Pond was available for fry rearing since our second pond was being used for holding cutthroat trout brood stock as part of a joint HSU-CFG brood stock development program. This resulted in a stocking density in Winter Pond #1 higher than previous rearing attempted in our Summer Ponds.

Fish:

Testing of the ponds was with surplus Eagle Lake rainbow trout available from the College of the Redwoods hatchery. These fry were reared in a redesigned recirculating tank and trough system (Allen and Couch Ibid). About 49,000 trout fry produced from the system were stocked in late spring and early summer 1989 into Summer Pond #1 (Table 1). These fish were fed Silver Cup pellets at about half the recommended rates (Klontz et al. 1985), reared until January 1989, then divided into equal numbers and released as "large" trout (8.0 fish per pound) into Winter Ponds #1 and #2 (Table 2). An additional 4,200 rainbow fry raised through the summer in the Arcata recirculating system were released into Winter Pond #1. These trout were relatively small (49 fish per pound), and are referred to as the "small" trout lot. The trout had eroded fins which aided in later identification. Trout in Winter Pond #1 were fed Silver Cup pellets, again at half the recommended rates, and trout in Winter Pond #2 were not fed and existed only on natural food organisms in the pond.

Table 1. Number and size of trout released into Summer Pond #1, 1989.

| Date Released | Number | Total Pounds | Number Per Pound | Vaccinated |
|---------------|--------|--------------|------------------|------------|
| May 22 | 24,200 | 46.9 | 500 1/ | No |
| July 1 | 24,400 | 59.2 | 413 | Yes |
| Totals | 48,600 | 106.1 | | |

1/ Average of four lots ranging from 428 to 630 fish per pound.

Table 2. Number and size of rainbow trout planted into 0.15 ha wastewater-seawater rearing ponds, AWWAP, January 1989.

| Winter Pond Number | Feed | Lot | Date Released | Number | Total Weight Pounds | Number Per Pound |
|--------------------|---------------------------------------|---------|---------------|--------|---------------------|------------------|
| 1 | Silver Cup Dry Pellets and Pond Fauna | "Small" | Jan 4 89 2/ | 4,164 | 85.5 | 48.7 |
| | | "Large" | Jan 23 89 | 4,420 | 552.5 | 8.0 |
| 2 | Unfed: (Pond Fauna only) | "Large" | Jan 23 89 | 4,428 | 553.5 | 8.0 |

2/ Vaccinated against vibrio

Trout in Winter Ponds were removed by pond draining and seining. Pumping down Winter Pond #2 began on April 22, followed on April 23 with only a partial draw-down due to mechanical problems with the pump. Final draw-down and fish removal occurred on April 24. Pumping down Winter Pond #1 was scheduled for May 5, but was only partially successful due to pump failure. The pump had to be repaired, delaying fish removal until May 11. Between May 5-11, bird netting had to be breached to allow hose access to the pond. Weak spots in pond netting and holes appeared frequently during this period. Repairs had to be made daily.

Trout removed from the ponds were placed in aerated holding tanks, and, after processing, were planted into Klopp Recreational Lake. The percentage of "small" and "large" trout released into Winter Pond #1 was determined by examining length-frequency graphs of fish planted and recovered (Figure 1). A sample of 124 trout from Winter Pond #1 were individually examined for the occurrence of beak marks (usually paired vertical bars on the caudal peduncle). Pond personnel were instructed to enter observations of birds within the pond netting into the daily log of pond operations providing records on heron activity. Incidence of beak marks on trout were also available from holding-tank mortalities, and from a mark-recapture study of trout as detailed later in this paper (Osburn 1989).

Some pond management problems occurred that impacted Summer Pond #1 operations

during 1988. A dike had been dug on the bank separating Winter and Summer Ponds #1, and filled with brush-bundles and alkali bulrush clumps. The ditch was used to study the possibility of reducing algal levels by pumping Summer Pond #1 water through the ditch. The ditch was lined with "hypolon" but seams became unglued and water leaked into the drained Winter Pond. This required adding makeup water to Summer Pond #1 from any available source, including city tap water, Humboldt Bay water, or available water from adjacent ponds. During the summer, water quality problems developed in Summer Pond #2, with water from Summer Pond #1 used to help alleviate the problems. As a consequence of these water pumping operations, there were times when the water in Summer Pond #1 was less than half its normal operating volume. Water additions produced a varying salinity regime during the course of trout rearing.

Terramycin-supplemented pellets were fed for five days beginning June 18, 1988 to combat a bacterial disease. Seine samples of trout to monitor length-weight were taken from the shallow, north end of Summer Pond #1. We found this procedure only sampled the smaller, presumably less aggressive trout- September 22: 10.3 cm mean fork length, 13.1 gm mean weight- 35 fish/pound; December 22: 9.65 cm mean fork length, 10.5 gm mean weight- 49 fish/pound. These data could not be used to show seasonal growth.

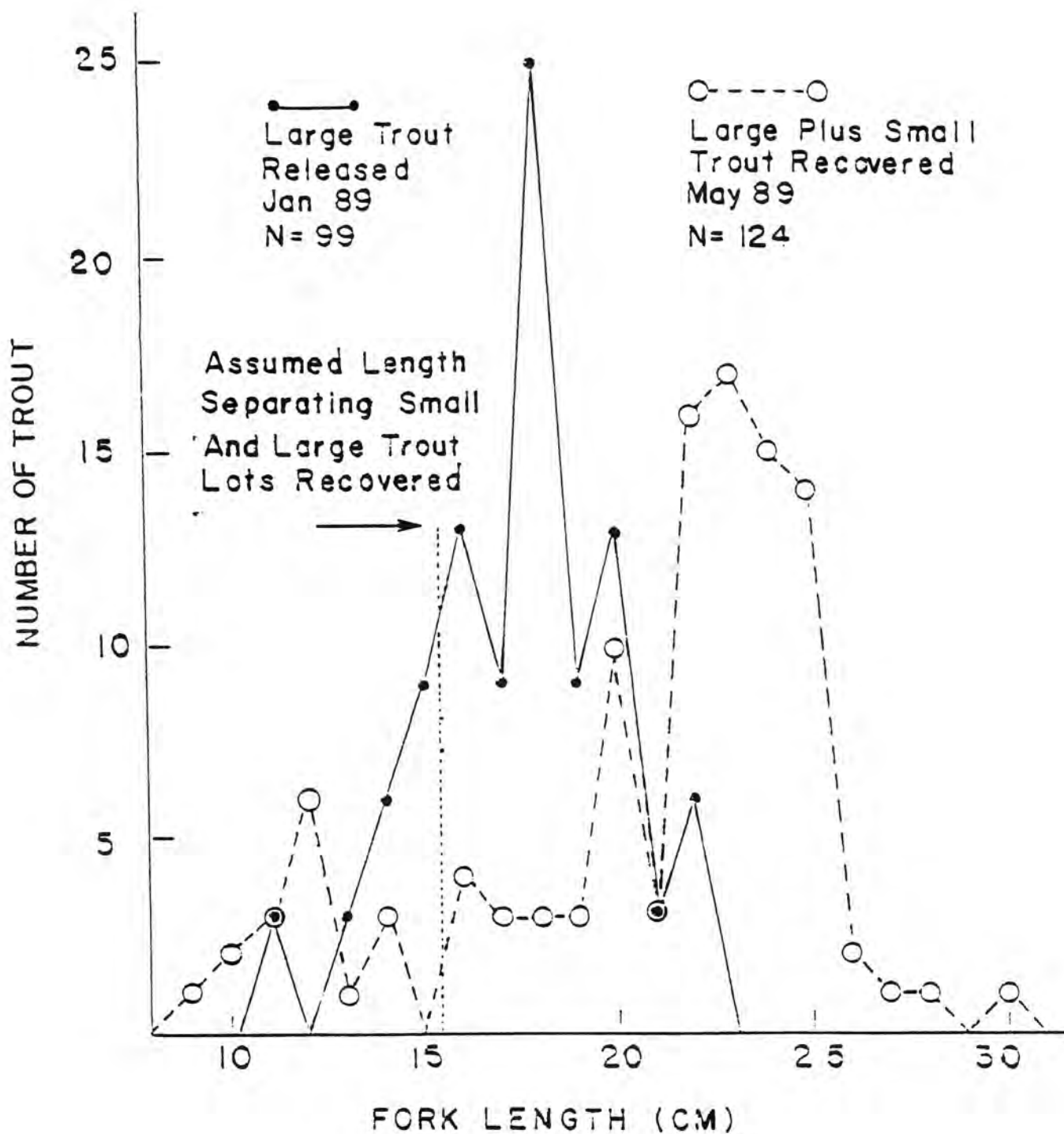


Figure 1. Length-frequency of trout sampled during draining of Summer Pond No. 1, January 1989, and of trout sampled during draining of Winter Pond No. 1, May, 1989.

WATER QUALITY

Five water quality parameters were monitored routinely. Reported here are conditions associated with Summer Pond #1 fry rearing only (Table 3). No readings were available during May 1988 during a period of personnel change. pH meter was inoperative from June 24 through August 1.

"Green water" dominated the pond from April through May (clarity less than 30 cm Secchi disc readings), but became somewhat clearer when a succession of brackish-water diatoms ("brown water") developed when pond salinities finally stabilized (15 - 17 ppt). "brown water" phase had clarities above 35 cm in Secchi disc readings.

Table 3. Summary of water quality parameters, Summer Pond No. 1, March 22 - October 31, 1989 (Values are mid-points of frequency intervals - I)

| Temperature (°C) | | Clarity (cm) | | Dissolved Oxygen | | pH Units | | Salinity (ppt) | |
|------------------|-----|--------------|-----|------------------|-----|----------|-----|----------------|-----|
| I | N | I | N | I | N | I | N | I | N |
| 13 | 1 | 10 | 1 | 3 | 1 | 7.0 | 10 | 1 | 3 |
| 14 | 8 | 15 | 13 | 4 | 4 | 7.5 | 8 | 3 | 1 |
| 15 | 14 | 20 | 1 | 5 | 13 | 8.0 | 12 | 5 | 5 |
| 16 | 12 | 25 | 3 | 6 | 4 | 8.5 | 3 | 7 | 6 |
| 17 | 5 | 30 | 6 | 7 | 1 | | --- | 9 | 5 |
| 18 | 2 | 35 | 2 | 8 | 7 | N | 35 | 11 | 3 |
| 19 | 3 | 40 | 13 | 9 | 6 | | | 13 | 9 |
| 20 | 2 | 45 | 1 | 10 | 3 | | | 15 | 9 |
| | --- | 50 | 2 | 12 | 2 | | | 17 | 6 |
| N | 47 | | --- | 13 | 1 | | | | --- |
| | | N | 42 | | --- | | | N | 47 |
| | | | | N | 45 | | | | |

Summer Pond temperatures remained below 17°C most of the summer and never exceeded 20°C. Two major oxygen regimes characterized summer rearing in 1988. The first phase, when "green water" and relatively low salinities were present, showed dissolved oxygen relatively high 7.8 - 13.2 ppm). A second phase, beginning in mid-July and running through at least to the end of October 1988, was associated with higher salinities. Oxygen levels were much lower (3.6 - 7.7 ppm, with most values around 5 ppm). Hydrogen ion concentrations were mainly from 7.7 to 8.5 during early low-salinity periods, then dropped to a 7.0 - 7.8 range during the late-summer period of elevated salinities.

Water quality data from November through January were routinely taken but became misplaced before duplicate copies were made.

RESULTS

Fish Production

Although survival in the fish barn was high, considerable mortalities occurred in both lots and ponds (Table 4). Large trout released into Winter Pond # 2 (unfed) suffered 29% mortality, showed no gain in size of fish, and produced less weight of fish than what was released (Tables 5 and 6). Small trout released into Winter Pond #1 (fed) suffered 85% mortality, produced less pounds of fish than planted, but doubled in individual size. In contrast, the large trout planted in Winter Pond #1 showed only a 13% mortality, doubled their individual size, and showed a net gain in total weight of about 460 pounds. Clearly, the natural carrying capacity of Winter Pond #2 could not sustain the 4,400 large rainbows planted. The feeding rate of pellets to the Winter Pond #1 population

produced fish of relatively good uniform size without grading (55% between 22 - 24 cm, Figure 1). Bird predation was

responsible for a portion of the 13% mortality found in large fish reared in Winter Pond #1.

Table 4. Number, net gain in pounds, and survival rates of domestic rainbow trout reared in Arcata wastewater aquaculture system, 1988 -1989 experiment.

| Stage | Rearing Facility | Number Produced | Net change in weight (pounds) | Percent Survival |
|------------------|------------------|-----------------|-------------------------------|------------------|
| Fry | Fish barn | 53,000 | + 64 | >95 |
| Fingerling | Summer Pond #1 | 8,848 | +900 | 18 |
| Yearling | Winter Pond #1 | | | |
| | Small trout | 626 | - 55 | 15 |
| | Large trout | 3,846 | +460 | 87 |
| | Combined | 4,472 | +405 | 52 |
| | Winter Pond #2 | | | |
| Large Trout | 3,130 | -156 | 71 | |
| Combined #1 & #2 | 7,602 | +249 | 58 | |

Table 5. Number and size of rainbow trout recovered from 0.15 ha wastewater - seawater rearing ponds, AWWAP, 1989.

| Yearling Pond Number | Date Recovered | Number | Total Weight Pounds | Number per Pound | Lot |
|----------------------|---------------------|--------|---------------------|------------------|---------------|
| 1 | May 5 - 11, 1989 | 626 | 31 | 20.0 | "Small" trout |
| | May 5 - 11, 1989 | 3,846 | 1,012 | 3.8 | "Large" trout |
| 2 | April 22 - 24, 1989 | 3,130 | 397 | 7.9 | "Large" trout |

Table 6. Percent occurrence of damaged fins, beak marks, pathology, and smolting in a sample of 124 individually examined trout for Winter Pond #1, May 10, 1989.

| Category | Percent |
|------------------------|---------|
| Missing or eroded fins | 22 1/ |
| Beak marked | 12 |
| Diseased | 6 |
| Smolt-like | 4 |

1/ Right pectoral - 13; all others combined - 9.

Fish Health

Only a few trout with any hemorrhaging were noted in the samples. Among 6 trout taken from the Winter Pond #1 fish trap on May 19, 2 trout had vibrio-like surface bleeding (Osburn 1989). In the random sample of 124 trout individually examined from Winter Pond #1, only 6% showed any pathological conditions (hemorrhaging, deformities, lesions, etc.) (Table 6). Bioassay with trout vaccinated and non-vaccinated against vibrio showed no treatment effect between the groups, and showed no apparent difference in survival from the large unvaccinated trout in the ponds.

Smolting

Trout utilized in this study were a domesticated strain for Eagle Lake, California, and therefore not expected to produce smolts. Since the trout were to be raised in saltwater and were to be fed in Summer Pond #1 and in Winter Pond #1, fish were expected to exceed the minimum size required for smolting in steelhead (roughly 17 cm) by the end of April or

early May when our ponds are harvested due to rising water temperatures. Sharp differences in both the peak time of entrance into smolt traps and in the average size of trout captured occurred (Table 7). The difference in trap catch between the two ponds during the initial period of Osburn's study was partly attributable to lack of installation of an entrance cone in the Winter Pond #1 smolt trap. The cone was added April 7. A period of increased trap catches occurred during the April 3-10 trapping period in both ponds. Out of the 21 days trapping in Winter Pond #2, on 19 days, trout were recovered that were greater than 15 cm mean average size. In contrast, there were only four days when the mean size of trout captured in Winter Pond #1 trap was greater the 15 cm, and two of these days occurred during a single period of trapping (April 11-16, Table 7). The data are intriguing to interpret. Possibly the smaller trout in Winter Pond #1 were trying to avoid predation by the larger fish, while the larger trout in Winter Pond #2 were trying to migrate due to lack of sufficient food or from the associated slimming process triggering DSM behavior.

Table 7. Catch of trout in smolt-traps operated in Winter Ponds #1 & #2, AWWAP, March 28 - April 20, 1989 (from Osburn 1989).

| Trapping and marking Period | Pond #1 | | Pond #2 | |
|-----------------------------|----------------|-----------------------------|----------------|-----------------------------|
| | Number trapped | Range in daily mean lengths | Number trapped | Range in daily mean lengths |
| March 28 - April 1 | 14 | 9.6 - 10.6 | 217 | 17.4 - 18.1 |
| April 3 - 10 | 28 | 8.6 - 14.4 | 69 | 14.8 - 18.1 |
| April 11 - 16 | 41 | 10.2 - 16.3 | 84 | 17.0 - 17.8 |
| April 17 - 20 | 22 | 12.0 - 16.4 | 54 | 14.7 - 17.4 |
| Combined | 105 | 8.6 - 16.4 | 424 | 14.7 - 18.2 |
| Mean | | 12.0 | | 17.1 |

We examined carefully trout sampled from the fish removed from Summer Pond #1 on May 4, 1989, for evidence of external morphological features characteristic of steelhead trout smolts. Smaller fish generally maintained the normal reddish band along the lateral line, strong parr marks, and darkish skin color of non-migratory trout. Bigger trout from the pond generally were much more silvery, with reduced lateral line coloration, and much fainter parr marks. Distal edges of fins of larger trout were slightly darkened but no fish were seen with the intense black edges we have observed on steelhead smolts produced from our ponds. Of the 124 trout examined intensively, only four showed external morphological facies that were "smolt like" (Table 6). These four fish ranged from 21.5 - 23.5 cm fork length. Condition factors of 0.95, 0.93, 0.92, and 0.91 showed typical slimming of a smolting fish (Winter Pond population C.F.: 1.23). We have had a few instances of increased smolt trapping associated with new moon (dark nights). New moon occurred on April 5 and full moon on April 20, 1989, neither date which could be associated with any increase in daily smolt trap catches. A sample of 20 large trout sacrificed for culinary purposes showed gonads still in the "thin string" stage. Since no physiological tests or enzyme analyses for smolting were made, many other explanations are possible for the trapping results on the two ponds and on possible smolting in these trout.

As the stock was considered non-migratory and was a non-native strain, informal state management suggestions favored release of all fish into Klopp Lake for recreational fishing. Thus, we probably missed an opportunity for the definitive test of the degree of migratory behavior remaining in this domestic trout strain by not studying their ability to survive in and return from the ocean to a release point.

Bird Predation- 1988

Heavy predation by cormorants on Winter Pond #1 has been reported previously (Allen and Couch 1988). Comparing survival of trout in a covered pond with an adjacent unnetted pond produced a slight improvement in survival rates but the causes of mortality were not easily partitioned (Table 8). Losses seemed to be associated

with factors other than birds. Our only study on night-time bird activities was completed during 1988 (Hopson 1988). He studied bird predation on Winter Ponds #1 & #2 in 1988 from a blind located between the two ponds. Observations for 1 - 3 hour periods were made using 8.5 x 44 power binoculars during the daytime, on clear nights, and under partial or full-moon nighttime conditions. On moonless nights, a model 221 Javelin light-intensifying spotting scope was employed. From February 27 to April 14, 1988 35 hours total observing recorded 40 herons and one kingfisher showing hunting behavior at the ponds. Of three different heron species recorded, the black-crowned night heron was most common. This was the only heron species recorded within the enclosed Winter Pond #1. Night herons were sighted seventeen times within the netted pond. Hopson observed night herons escaping from within the netted pond twice by prying themselves through the seam connecting the netting to metal fence posts along the north side of the pond. Night herons were observed twice unsuccessfully attempting to penetrate the netting from outside the pond. No night herons were observed capturing fish within the netted pond, but in the unnetted pond on April 3 and 7, observations a capture rate of .27 fish per hour (4 fish consumed over 14.5 hours of observation). Hopson felt that night herons gained access both by utilizing any loosely connected seams or by actually severing nylon mesh. Many of the 30 holes which he repaired and marked using colored plastic tape were frayed or serrated as would be expected from the gripping edge of the night heron's bill. Most sightings of hunting herons were in the early evening and early morning hours (Figure 2), with night herons representing 83% of all heron sightings. Hopson noted that the 7 - 11 ppm maximum hunting activity correlated with increased surface feeding by fish in the ponds at sunset. This is congruent with Watmough (1978) who suggested that increased fish activity at night was responsible for higher fish intake by night herons feeding at night than during the day. Hopson did not attempt to estimate a possible total consumption of fish removed from the ponds in 1988. River otters weren't sighted in either Winter Pond during the study, although scat and tracks were evident in closely adjacent areas.

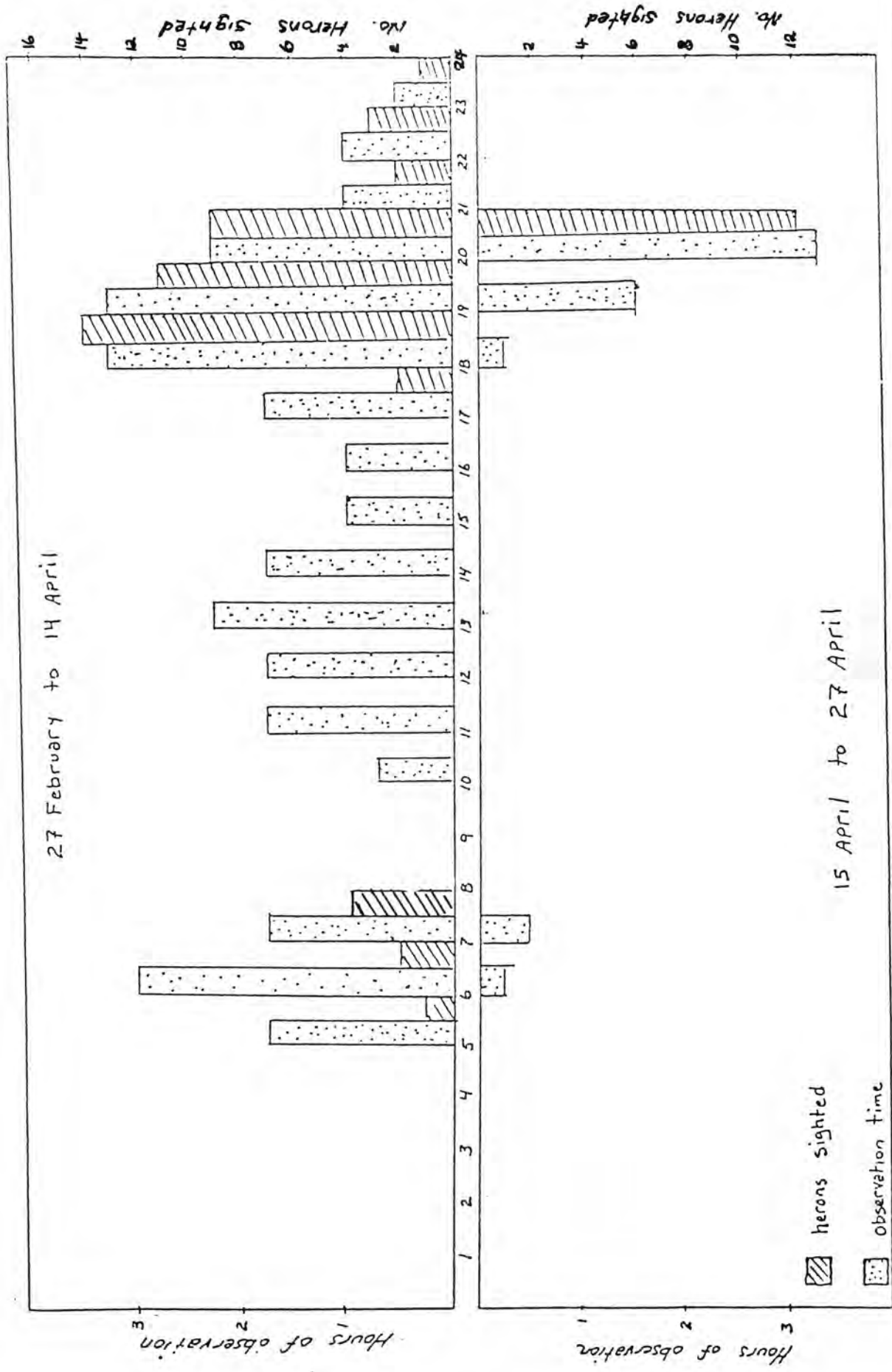


Figure 2. Number of hours of observation and number of herons sighted by one-hour periods, AWWAP, February-April 1988. (Period 27 February-27 April includes combined data from Winter Pond No. 1 (covered) and Winter Pond No. 2 (not covered); Period 15-17 April are data from Winter Pond No. 1 when a small panel of pond netting was breached for experimental purposes). (Winter Pond No. 2 drained 15 April; Winter Pond No. 1 29 April) (from Hobson 1988).

Table 8. Comparison of survival rates of juvenile trout and salmon reared in Winter Ponds and probable cause of mortalities, AWWAP, 1987-1989.

| Year | Species | Percent Survival | Loss Factor (s) | Pond Netted | Species | Percent Survival | Loss Factor (s) | Pond Netted |
|------|-----------------|------------------|-----------------------------------|-------------|-----------------|------------------|------------------|-------------|
| 1987 | Coho | 12 | Cormorants | No | Coho | 9 | Low D.O. | No |
| 1988 | Coho | 55 | Probably H ₂ O Quality | Yes | Steelhead Trout | 37 | Hérons; Low D.O. | No |
| | Steelhead Trout | 42 | | | | | | |
| 1989 | Rainbow Trout: | | | | Rainbow Trout: | | | |
| | Small | 15 | Cannibalism | Yes | Large | 71 | Starvation | Yes |
| | Large | 87 | Minor loss to Multiple causes | Yes | | | | |

Bird Predation- 1989

No personnel were available for night-time observations of bird predation in Winter Ponds in 1989. Assessment of mortality factors used survivals in trout held in cages in the pond and through a study of heron attacks as shown by beak marks on fish samples.

A single cage floated in each of the two Winter Ponds was utilized as a predation and survival control for the 1989 experiment. Sixteen trout were placed in each cage, with one cage employing vibriovaccinated fish and the other unvaccinated. Bioassay fish in Winter Pond #2 were transferred on April 22, 1989 to the control cage in Winter Pond #1. The two lots were kept separate by a divider placed in the cage. On May 4 fish from both cages were counted, measured for length and weight,

and examined for beak marks (Table 9). Data from the control were subject to error. First, more fish were recovered than placed into one side of the pen, although this might have been a mistake made when transferring fish. Second, there was a high rate of beak-marked trout (19%). Night-crowned herons undoubtedly used the cages as feeding platforms during the 12 days when the cage with the two lots of fish were in Winter Pond #1. Fish could have died from heron attacks during this period or during previous months of rearing. Combining both vaccinated and unvaccinated trout, total mortality in bioassay cages was only 16% but cannot be partitioned with confidence between water quality, disease, or predation. Losses strictly due to bird predation were some fraction of the 16%, shared with other causes such as fish health, water quality, or perhaps other unrecognized factors.

Table 9. Results of cage bioassay of Winter Ponds #1 & #2 using 16 vaccinated and 16 unvaccinated trout per cage, AWWAP, January - May, 1989.

| Pond Number | Vaccinated | Number Recovered | | | Percent Beak-Marked | Mean Size | |
|-------------|------------|------------------|-----------------|------------------|---------------------|-----------|--------|
| | | Live | Dead | Total | | L (cm) | W (gm) |
| 1 | Yes | 12 | 1 ^{1/} | 13 | 23 | 17.2 | 77.0 |
| 2 | No | 15 | 2 ^{2/} | 17 ^{3/} | 12 | 16.9 | 68.5 |

1/ Decomposed

2/ Fresh dead

3/ More than fish reportedly placed in cage. Counting, transfer, or fish migration error.

Beak Marks

Trout surviving night-crown heron attacks can be identified by the presence of beak marks. Beak-marked trout in 1989 could be compared from four samples:

1. Smolt trap catches (Winter Pond #1- 105; Winter Pond #2- 424).
2. Dead trout recovered from bottom and brush-bundle reefs after draining Winter Pond #1, May 4-5, 1989- 40.
3. Sample of trout from Winter Pond #1 taken during draining of Winter Pond #1, May 5, 1989- 124.
4. Holding tank mortalities trout removed from Yearling Pond #1- 308.

Trout in smolt traps were counted on 21 days between March 28 and June 20, 1989 (Osburn 1989). Captured trout were fin-marked according to four periods of capture, and all captures during a time period were released to a different corner of the Winter Pond in which the smolt trap was located (Table 7). From 105 trout captured in the smolt trap in Winter Pond #1, only 6 marked trout were recaptured (6%), while 90 recaptures were recorded from 424 marked trout released (21%) in Winter Pond #2. Among captured Winter Pond #2 trout, there were 38 fish which were recaptured at least twice (three

missing fins). Three "trap happy fish" were recaptured three times (four missing fins), but these were from 54 fish released during the last marking period that were returned to the pond in the corner adjacent to the smolt trap. Of six recoveries in Yearling Pond #1, all were from trout released during initial trapping, with the first recovery on April 4 and the last on May 20 when the study was terminated. Osburn only recorded beak marks during the first week of his study. Trout appeared particularly susceptible to bird predation on entering the Winter Pond #2 smolt trap (Table 10).

During the period from April 22 to May 4, personnel were asked to note any egregious bird predation events on Summer Pond #1. On May 2, on arriving at 9 A.M. at the ponds, fish culture staff observed three black-crowned night herons within the pond enclosure. Two of the birds were flushed from the enclosure through the pond gates. The other bird could not be flushed out. At 3 P.M., 12 dead trout were counted either on the pond bank or floating on the pond surface. At 6 P.M. the heron was not present inside the netted pond. A survey of Winter Pond #1 pond bottom on May 4 and of the brush-bundle reefs on May 5 recovered 40 fish, of which a minimum of 11 mortalities appeared directly released to heron feeding (Table 11). The data showed no obvious select of attacked trout by size by the herons. We were not able to record data on the actual size of trout consumed.

Table 10. Percent of trout with beak marks captured in Winter Pond #2, March - April, 1989.

| Date Captured | Number Caught | Number Beak-Marked | Percent Beak-Marked |
|---------------|---------------|--------------------|---------------------|
| March 28 | 131 | 4 | 3.1 |
| 29 | 58 | 5 | 8.6 |
| 30 | 14 | 4 | 28.6 |
| April 8 | 8 | 1 | 12.5 |
| Total | 211 | 14 | 6.7 |

Table 11. Status of dead trout recovered from Winter Pond #1 pond bottom AWWAP, May 4, 1989 (18 fish) and from brush-bundle reefs May 5, 1989 (22 fish).

| Category | Number | Size (cm) | | |
|---------------------------------|-----------|-----------|------|------|
| | | Min. | Mean | Max. |
| Freshly regurgitated | 4 | 17 | 20 | 23 |
| Fresh dead | | | | |
| Carcasses with beak marks | 7 | 13 | 21 | 26 |
| Carcasses without visible marks | 7 | 16.5 | 20 | 24 |
| Decomposing carcasses | 14 | 9 | 19 | 24 |
| Skeletons | 8 | - | - | - |
| Total | 40 | | | |

Table 12. Percentage of trout removed from Winter Pond #1 showing evidence of attacks by bird predators for the presence of beak marks, AWWAP, May 5 - 10, 1989.

| Sample Source | N | Beak marked | | Size (cm) | | |
|--|-----|-------------|---------|-----------|------|------|
| | | Number | Percent | Min. | Mean | Max. |
| Holding tank mortalities | 308 | 10 | 3 | 16.5 | 20 | 24 |
| Random Sample held for length-weight studies | 124 | 15 | 12 | 14 | 23 | 30 |

Two additional samples of Winter Pond #1 beak-marked fish varied between 3% and 12% (Table 12). The value found in the random sample was based on examination of live fish while the value determined on mortalities was from fish dead for about 48 hours.

DISCUSSION

Hopson (1988) saw few herons inside Winter Pond #1 during night-time observations, with only 26 beak-marked fish noted in combined smolt-trap catches and recoveries on pond draining (0.087% beak-marked). This low incidence correlated with Hopson's lack of observation on fish being consumed by herons inside Winter Pond #1 in 1988. The corresponding percent of beak-marked

trout occurring in recoveries from the unnetted Winter Pond #2 in 1988 was 0.012%. He recorded on Winter Pond #2 a capture rate of .26 fish per hour from a capture efficiency of one fish per 4.5 strikes. We unfortunately do not know how many strikes actually left beak-marks on fish, the mortality rate of struck fish, or the average time until death after attack, all of which are needed to estimate total losses based on beak-mark analysis. Hopson's study, however, did not suggest any serious losses due to bird predation in 1988.

Complete netting of Summer Ponds in 1989 was correlated with a survival of larger trout of 71 - 89%, a much reduced rate of mortality over 1987 and 1988 (Table 8).

Bird predation was eliminated as a source of mortality except for occasional black-crowned night herons. We have a hypothesis that only a few birds learn quickly to penetrate pond netting and the pond becomes a feeding space defended by individual birds. Some indication of this is from the fact that entries into Summer Pond #1 and Winter Pond # 1 involved only single birds (Tables 12 and 14), with observations of two birds inside the netting reported only once in either pond. Following extreme aggressiveness of herons in late October and early November, pond personnel on November 12, 1989, hung monofilament gillnet across the south end of Summer Pond #1. One night heron

became tangled in the net on November 25, was captured, head feathers spray-painted red, and released. A second bird was captured, spray painted, and released on November 28. One marked bird was recaptured on November 29 and another on December 3. No additional sightings of night herons inside the pond were recorded through the remainder of December and through to January 23 when the pond was drained. Although only a few individual birds may be involved in fishing a pond, if fish densities are high, the birds will tend to gorge themselves. Early in June 1989 in Summer Pond #1, a night heron flushed from the pond regurgitated over 100 small coho.

Table 13. Summary by month of number of days when black-crowned night herons were found inside enclosed Summer Pond #1 as recorded in daily log of pond operations, AWWAP, April - December 1988.

| Month | Number of days bird recorded | | Repairs to Netting reported |
|-----------|------------------------------|-----------|-----------------------------|
| | One bird | Two birds | |
| April | 0 | 0 | 0 |
| May | - | - | - |
| June | 3 | 0 | 5 |
| July | 0 | 0 | 0 |
| August | 0 | 0 | 2 |
| September | 0 | 0 | 3 |
| October | 3 | 0 | 0 |
| November | 7 | 1 | 2 |
| December | 1 1/ | 0 | 0 |

1/ Grebe

Table 14. Summary by month of number of days when black-crowned night herons were found inside enclosed Winter Pond #1 as recorded in daily log of pond operations, AWWAP, January - May 1989.

| Month | Number of days bird recorded | | Repairs to Netting reported |
|----------|------------------------------|-----------|-----------------------------|
| | One bird | Two birds | |
| January | 1 1/ | - | 0 |
| February | 6 | 1 | 3 |
| March | 5 | 0 | 0 |
| April | 2 2/ | 0 | 0 |
| May | 0 | 2 3/ | 0 |

1/ Egret

2/ One kingfisher and one cormorant

3/ one inside pond and one sitting on netting

Although we cannot present the results of any specific study, our general experience is that herons (great blue heron, great egret, and black-crowned night heron) are all quickly attracted to the ponds when fish become available for capture. Increased availability may come from concentration of stressed or sluggish fish along pond banks resulting from disease or poor water quality, or from smolts cruising pond banks during downstream migration periods. Herons are very quick to discover any gaps in smolt trap covers, and day-time feeding herons (great blue heron and white egret) are almost impossible to frighten away from ponds during such periods. We suggest that undue concentrations of herons can actually be used as an early indicator of fish stress, and thus for possible early initiation of appropriate management action to alleviate fish losses.

Other species of birds that have taken fish from unprotected ponds include the belted kingfisher (*Megaceryle alcyon*), double-crested cormorant (*Phalacrocorax auritus*), caspian tern (*Hydroprogna caspia*), osprey (*Pandion haliaetus*), and six species of gull (*Larus sp.*). Netting has effectively eliminated fishing by these species except on those occasions when pond gates are left open and birds walk into the enclosure.

MANAGEMENT

Bird predation should be a minimal concern in the future with both ponds now completely covered. Night herons can be expected to penetrate enclosures but the incidences can be controlled by instituting daily patrols to repair holes and to insure gates are not left open overnight. Overhead netting on Summer Ponds will be raised to allow repair of seams from a boat which is now not possible. Should heron predation continue, probably scare devices by intermittent light flashes or noise during a 3-hour period after sundown would make the ponds even more unattractive to the birds. A dog within the pond enclosure is highly effective against day-feeding herons but has not been tested against night herons. Predation by land otters can be expected if precautions are not taken. Winter Ponds are now ringed with a one-strand electric wire placed at the outside base of enclosure walls. Extra-heavy wire mesh tops are to be constructed for all outside holding tanks.

Additional attention will be given to maintaining high aeration rates, particularly in Summer Ponds, as well as developing a flexible back-up aeration unit. Since our mortalities were associated with both high and low dissolved oxygen levels in the Summer Pond during 1988, other critical parameters such as ammonia will have to be monitored routinely to more accurately partition presumptive causes of mortalities.

Whether a "green water" system or a "clear water" system resulting from *Enteromorpha*-dominated ponds is more productive remains unclear. Winter Pond #1 (feed) contained considerable pond fauna on draining as compared to Winter Pond #2 (no feed). Recent pond experiments have involved seeding ponds with *Enteromorpha* but have not been successful in establishing a dominating plant population. The oxidation pond waters may have become less fertile due to upgrading of the primary treatment units of the Arcata sewage treatment facilities. Such water when mixed with saltwater could be providing a less suitable substrate for development of *Enteromorpha* which favors organically-enriched waters.

A much improved (61%) and more characteristic Summer Pond survival rate has already been obtained with coho salmon fry reared from June 13 to November 18, 1989 (13,000 fry planted at 157 fish per pound and 8,000 fingerlings recovered at 48 fish per pound). Rogue coho (75 fish) were 5.4 fish per pound in weight and were released to Humboldt Bay via South Pond. Winter Ponds #1 and #2 each received 4,000 coho fingerlings to again test pond netting. Smolts from the 1990 Winter Pond study are scheduled for imprinting in South Pond. The release site will be under a tidal flushing regime, with a continuous addition of effluent from the Arcata marsh and wildlife sanctuary. Dissolved organic compounds in the effluent should provide an imprinting substrate. Smolts will be forced to migrate through the pipe connecting the South Pond into Humboldt Bay. Marsh effluent is planned for use as attracting water for returning adults.

ACKNOWLEDGEMENTS

We are indebted to the College of the Redwoods aquaculture program for providing trout fry used in testing the 1988 redesigned recirculation system and availability for testing covered ponds in 1989. Prairie Creek County Fish Hatchery was our source of coho salmon eggs. Humboldt State University students enrolled in a class for wastewater aquaculture provided much needed personnel required for removing trout from ponds during the spring of 1988 and 1989. Vaden Janzen, aquaculture trainee, contributed maintenance and construction skills, and assisted in feeding fish. Delores Neher, California Cooperative Fisheries Research Unit, prepared the final copy of this report.

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IDEAS, INNOVATIONS, AND GADGETS

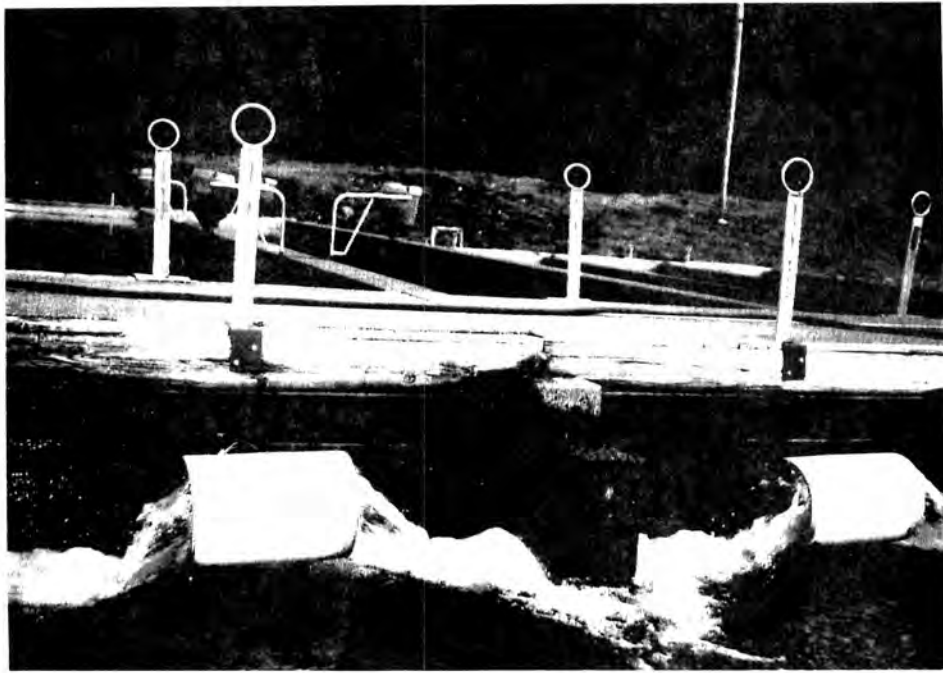
Ulf Rasmussen
Washington Department of Wildlife
South Tacoma Hatchery
7723 Phillips Road SW
Tacoma, WA 98498 (206) 964-7267

The presentation was an attempt to share the ingenuity of our colleagues at various facilities throughout the State of Washington. Good ideas often pop up in response to individual problems and sets of circumstances. Keeping in mind that hatcheries differ, things were shown that

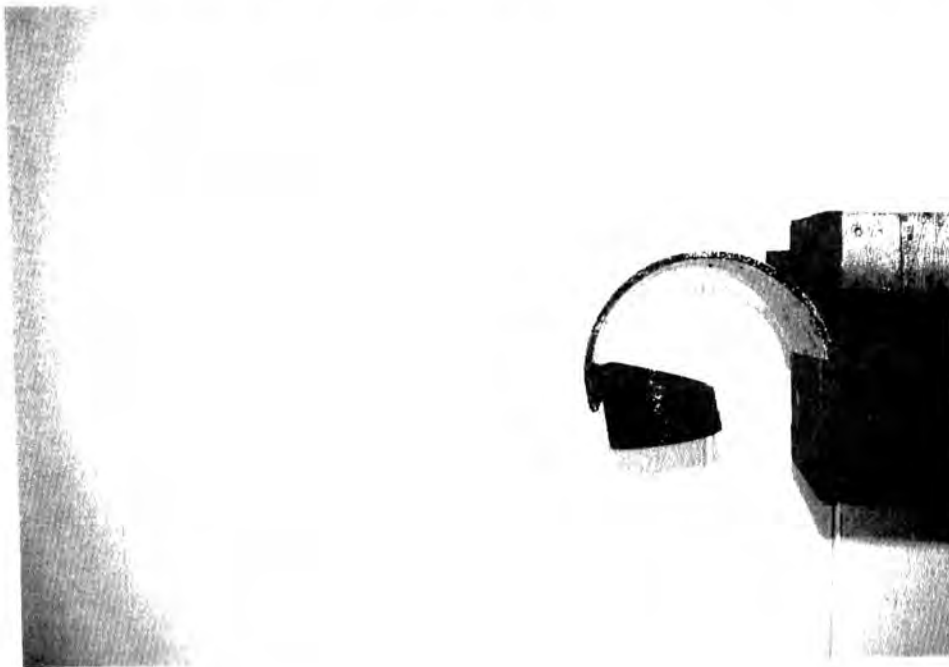
were of no use to some but could prove valuable to others. Not all of the ideas/innovations were brand new but all have made life easier and more productive for the personnel involved. The presentation given consisted of 70 slides. The following are selected highlights.



Boot drier- easy assembly of ABS plumbing, holds chest waders, the air intake is underneath the ceiling where it is warmer.

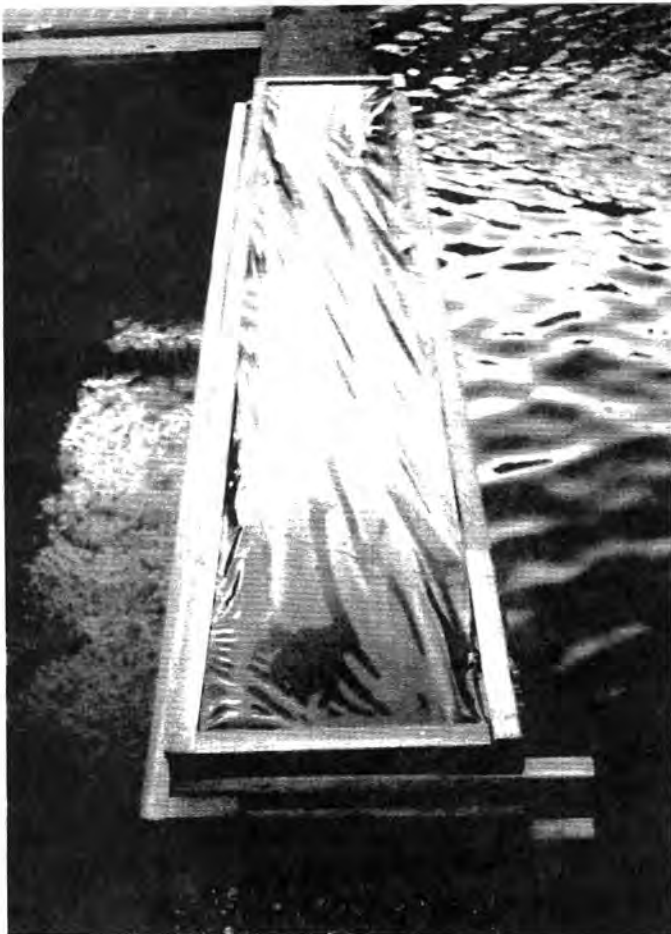
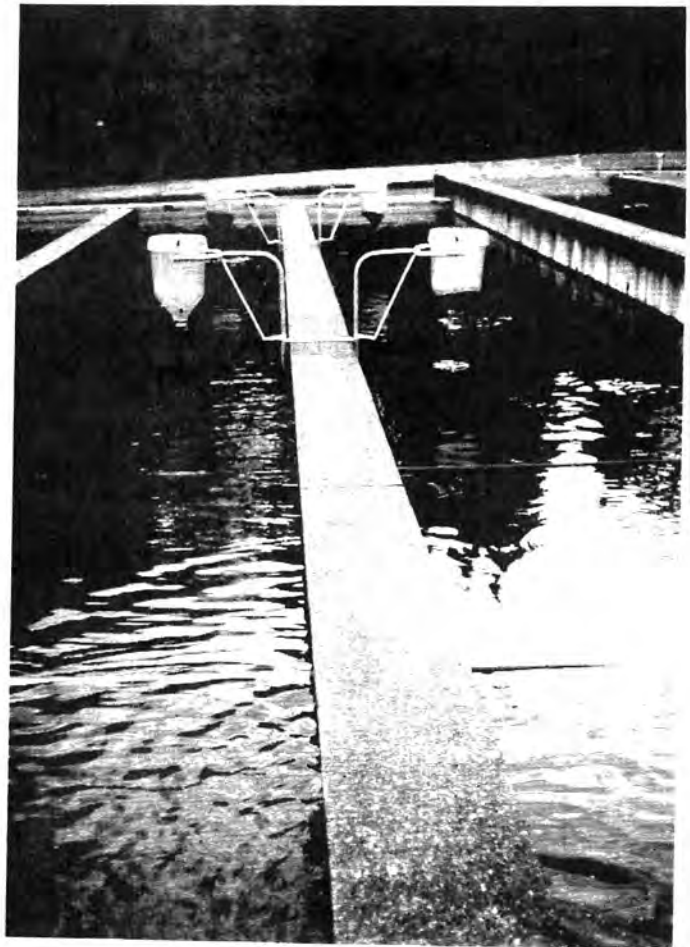


A short piece of 4 inch pipe directs the water towards a section of 10 inch PVC sewer pipe. Fish will jump towards water entering the pond but cannot make the 90 degree turn into the 4 inch pipe. Notice how close the deflector is to the level in the pond. In shallow raceways, a small increase in depth will give you a significant increase in rearing space. For example, if you add 6 inches to an average depth of 24 inches, volume will increase 25%. Another benefit, this deflector will not plug up with debris.



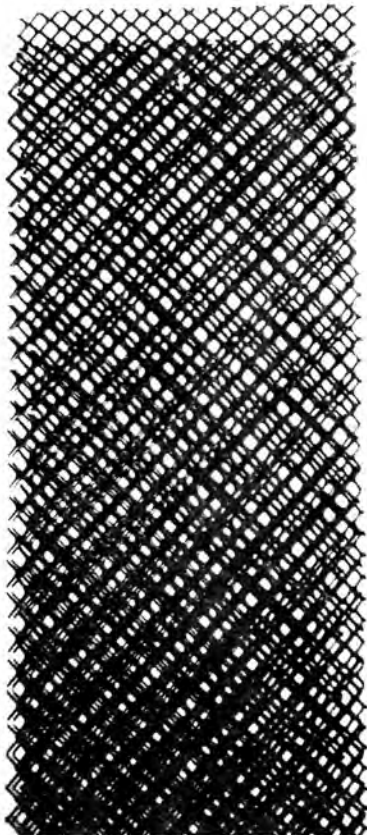
Notice that the lower end of the coupler has been removed. Depending on head and flow, this may be necessary to avoid back pressure and reduced flow.

Two baffles 8 feet apart in the upper end make on-third of this 10X80 raceway self cleaning. Savings in time needed at pushing the broom is closer to half. (No piles of poop under the intake.) Also, the ponds cannot be drawn down too far because your employee is done cleaning before he has had the chance. While acknowledged to be better for the fish, this is also less stressing on many managers.

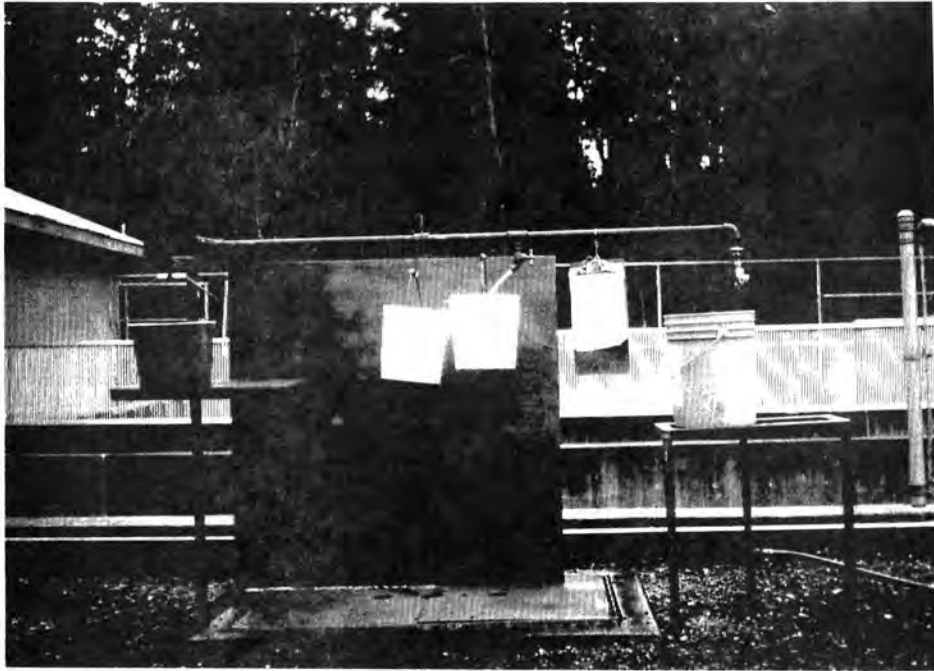


Baffles do not have to be expensive. This model consists of 4 mil black plastic, 2X2 lumber and lath. The legs make them easy to wedge in at the correct depth. The depth (actually the distance between the baffle and the bottom of the pond) depends on turn-over. Three to four inches seems to be an average.

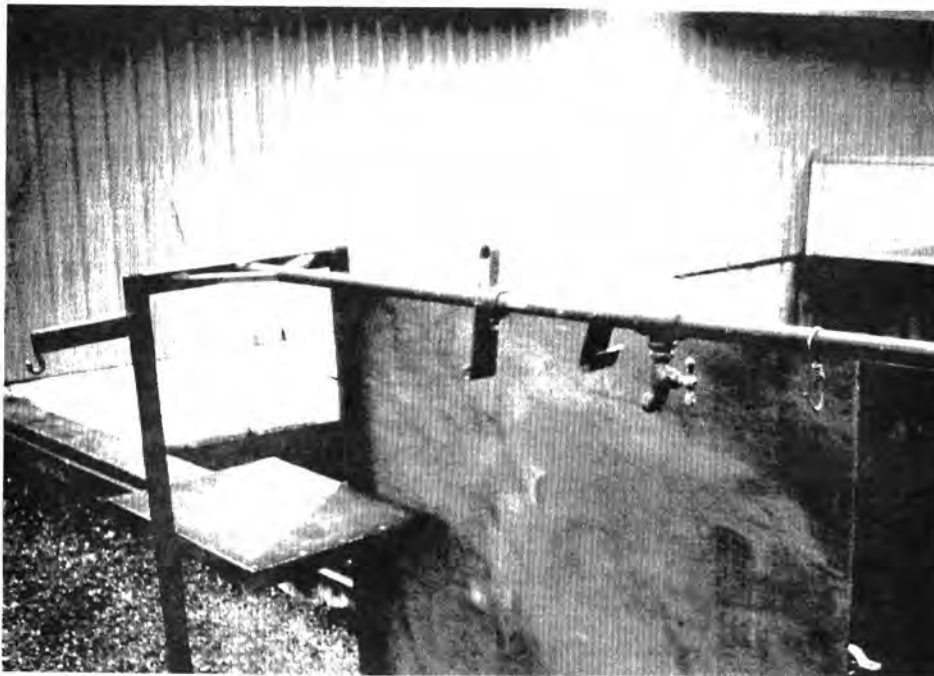
This facility, which is a warm-water station, is hatching an additional half a million steelhead after somebody thought of doing it under covers in outside intermediate raceways. It has the potential of hatching another half a million. Using materials on hand, the staggered baskets are supported by rebar which rests on cinder blocks and bricks. It is necessary to elevate the baskets over the substrate to avoid suffocation. No up-welling is required as long as the eggs are kept to 1-2 layers. Note, the cater coming in has to be dampened or the eggs in the upper baskets may pile up and smother. Also, the fly screen in the lower end cleans easily with a garden hose while the water is drawn down.



Substrates have proven beneficial to many hatcheries. This is a sandwich of fine mesh between pieces of coarse mesh, which creates more niches.

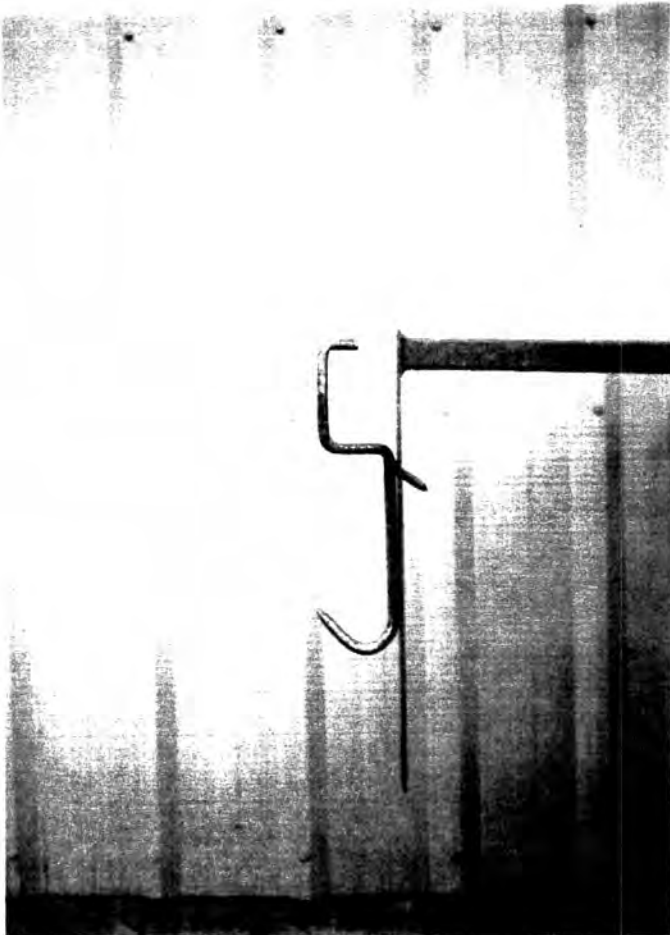
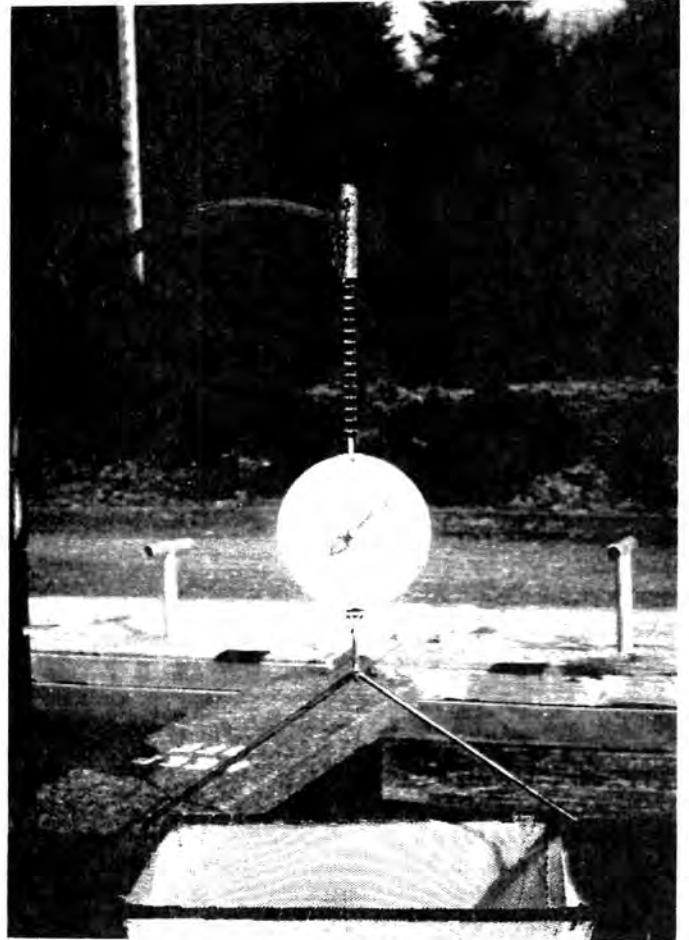


Spawning in a sanitary fashion; virus-free water is brought to the spawning area through a garden hose. The disinfecting station carries the water through its plumbing. Hand disinfectant to the left; iodophore buckets to the right; a clip board for noting the onset of hater hardening in iodophores. Of the two center buckets, the bucket to the left contains fertilized eggs, while the bucket to the right provides a continuous reservoir of virus-free water. There is also a faucet over the iodophore buckets. Using the set-up is fast, efficient, and easy on the back.



Details. The hook to your left holds the hand disinfectant. Underneath is a platform for egg buckets. The hook next to the faucet hold the reservoir for virus-free rinsing water. The remaining hook is used for hanging the fertilized egg buckets. Notice that this hook is lower than its neighbor for easy filling. Also, it moves on the pipe and swivels for easy draining. Excess sperm , ovarian fluid, and rinsing water all drains into a dry well underneath the rinsing station.

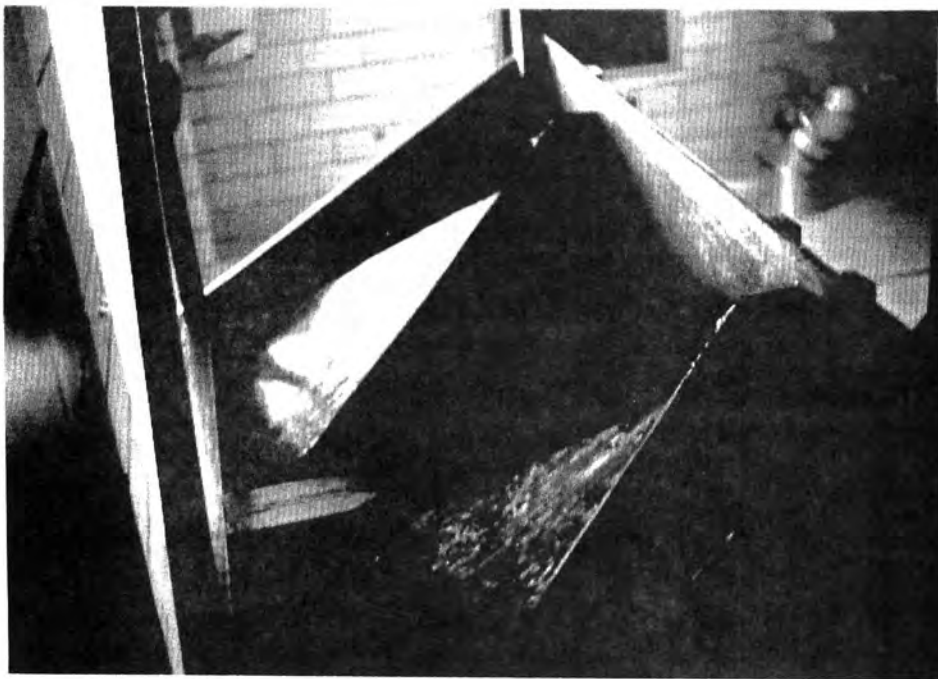
The scale that does not twist, regardless of how windy it is. A chain runs inside a pipe. It can be adjusted at any level and at any angle by a pin inserted through holes drilled into the pipe.



A spawning hook may save a person ordinarily used for hold the fish. The hook slips into elongated holes that have been drilled down at a 45 degree angle into a piece of flat iron. (Shown from the side.) A dozen holes one inch apart will allow you to position the vent of fish of various lengths exactly at the colander.

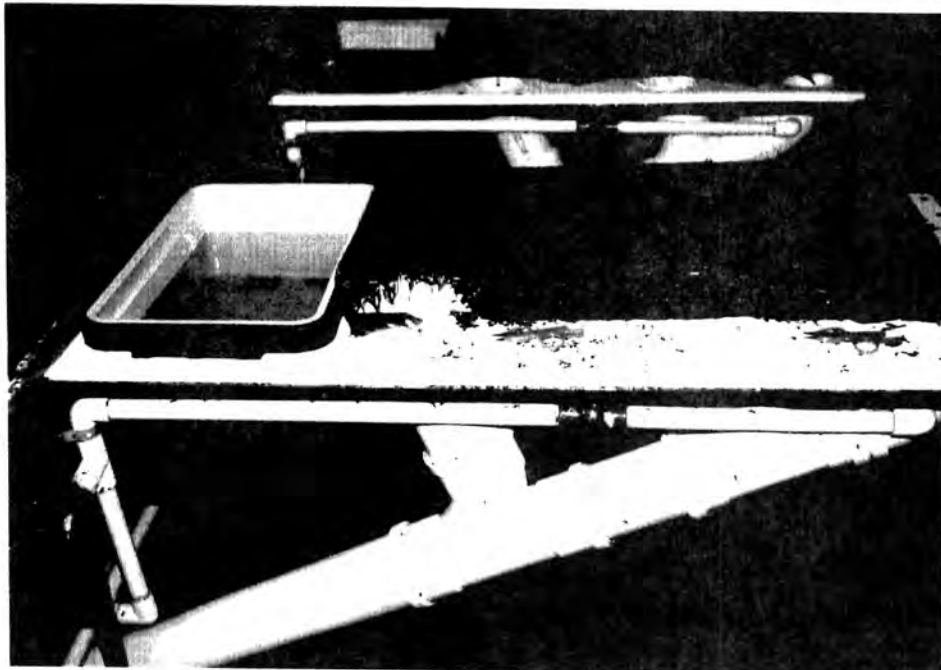
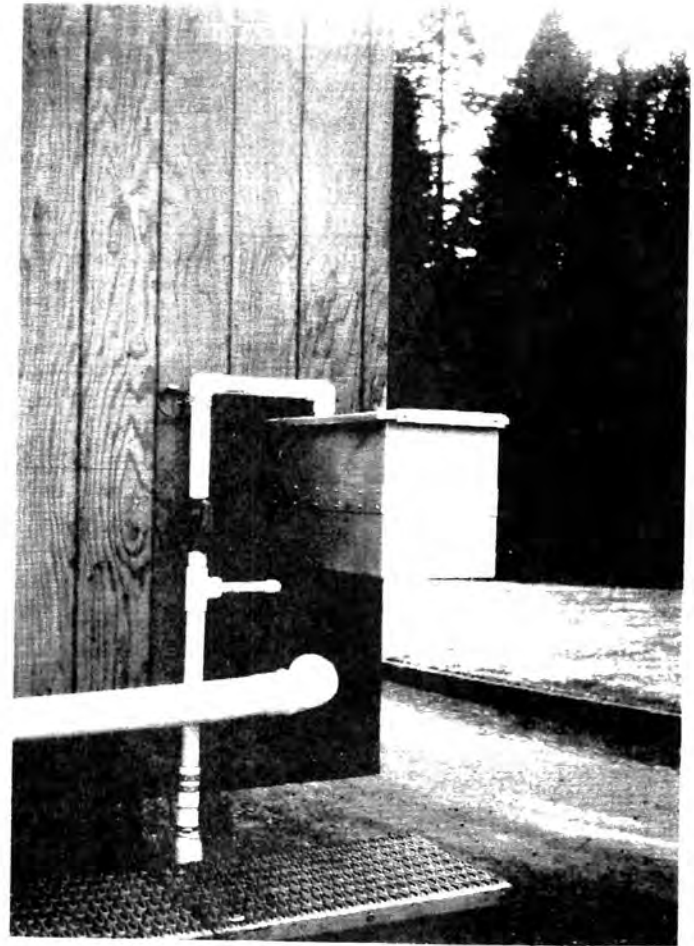


The feed bin is a big help if you do a lot of feeding by hand and need to keep track of the feed for different groups of fish. The bin should be tall enough so that most people can stand up straight at the scales.



To avoid dead spots in the corners, the bottoms of each compartment may consist of three pieces of plywood that are glued and screwed together

Details from a home made transportable marking shed: The deep tank consists of two old aluminum troughs that are joined together by using pop rivets. You may laugh, but it was free.



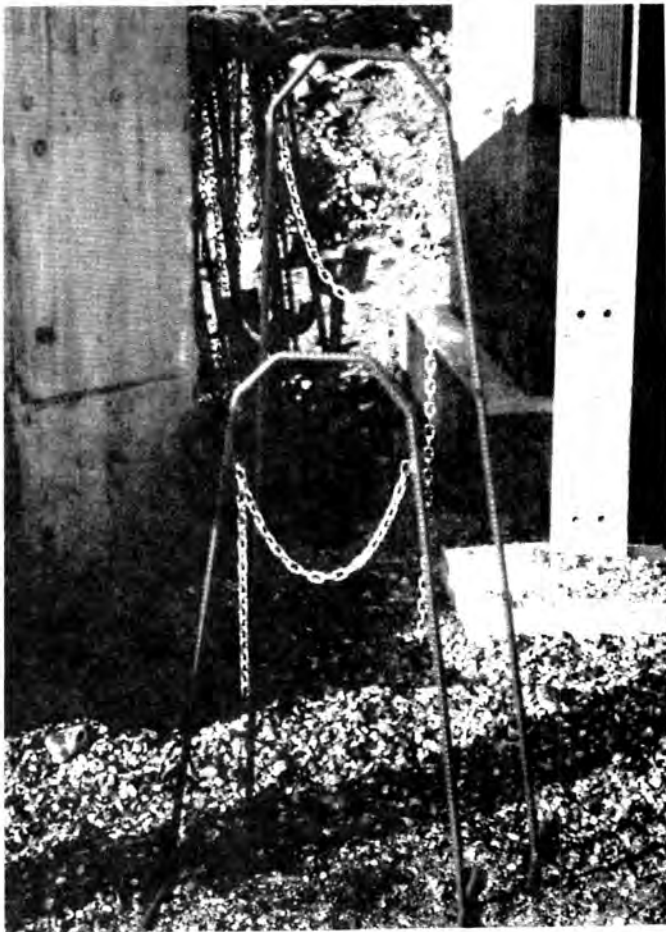
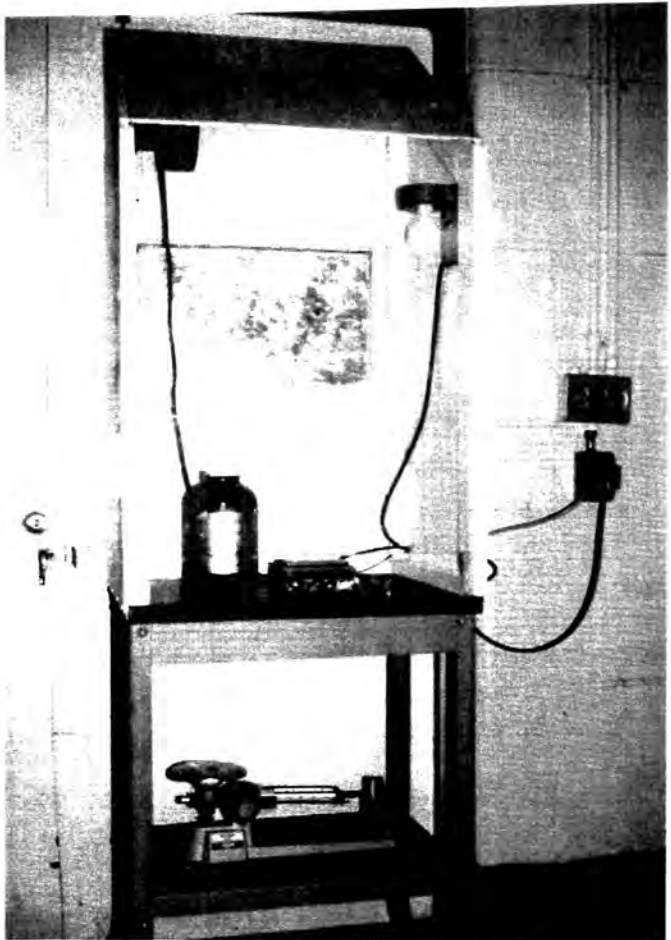
The deep tank runs to the left. Each clipping station has an anesthetizing tub which provides fish for three people. The tub is a Coleman Cooler. The insulation helps when it is warm.

An excellent solution for avoiding overheating while clipping in hot weather. The MS-222 is contained in a garbage can that fits snugly inside the coils. Notice the simple but effective net holder.



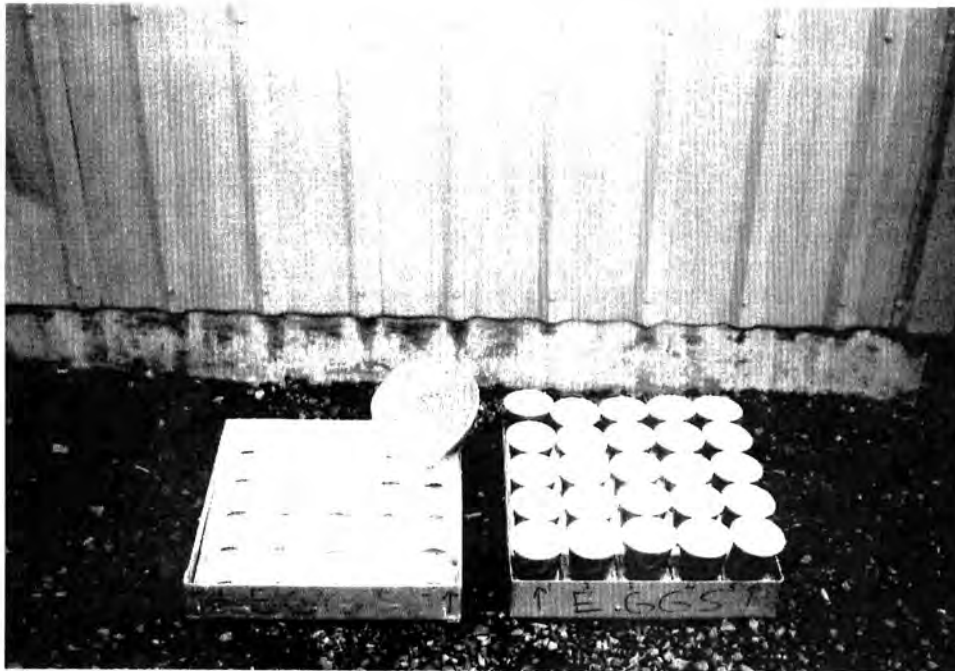
To appease a pathologist; this movable disinfecting cart carries two 18 inch brushes with 12 foot handles from pond to pond. One brush is disinfected while you use the other.

Fish hatchery workers handle several dangerous chemicals. The ventilated hood is used everywhere else, so why not by us? This model was built from junk- a discarded router table, part of a formica table, a sliding plexiglass window, an old light fixture, and the fan from a fireplace insert. The only new part that is new is a multiple switch that makes you turn on the fan when you want to use the electronic scale.



The most elegant pipe support you have ever seen?

We all know the person who cannot lift the feed sack high enough. The solution is a piece of pipe attached to the end of the first roller.



An increasing concern; genetic health. The "5 X 5" spawning consists of splitting the eggs from each of five females into five different cups, then fertilizing the cups with sperm from five different males. Five males and five females will give you 25 male-female combinations. The fertilized eggs are put together and raised as a normal 5-fish pool. Time needed may be 8-9 minutes as compared to 4-5 minutes per pool when you are not spawning brood replacement.

HISTOLOGIC EFFECT OF CODED-WIRE TAGGING CHUM AND COHO SALMON FRY

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ABSTRACT

Chum salmon fry (*Oncorhynchus keta*), average weight 0.58 grams, and coho salmon fry (*Oncorhynchus kisutch*), average weight 0.23 grams, were tagged with half-length (0.5-mm long) coded-wire tags. Histologic examination revealed mainstem olfactory nerve damage and/or placement of tags into the frontal lobe of the brain in 18% to 37% of the fish examined. Such damage and placement should be of particular concern because of the well-documented role olfaction plays in salmonid behavior and the low regenerative capacity of nervous tissue.

INTRODUCTION

The need to identify large numbers of fry and emergent migrating Pacific Salmon has increased substantially over the past few years. To date, the preferred method for identifying these small fish is the half-length coded-wire tag. Reports of successfully tagging pink salmon (*Oncorhynchus gorbuscha*) as small as 0.25 grams/fish have been described (Thrower and Smoker 1984), yet the half-tag appears most satisfactory when fish are approximately 1 gram or larger (Moberly et al. 1977; Blankenship 1981; Opdycke and Zajac 1981). Data supplied by the Pacific Marine Fisheries Commission, Portland, Oregon (1989), documents that between the years 1980 and 1988, over 5.3 million Pacific Salmon were coded-wire tagged at a weight of less than one gram. Morrison and Zajac 1987 demonstrated mainstem olfactory nerve damage in chum salmon that were half-tagged at an average size of 0.67 grams. This finding was of particular concern given the documented role olfaction plays in salmonid behavior (Hoar and Randall

1971; Hasler and Scholz 1983; Doving et al. 1985). This paper reports additional information supporting a potential problem when coded-wire tagging very small salmonids.

METHODS

Chum salmon, average weight 0.58 grams (range 0.54 - 0.62) and coho salmon, average weight 0.23 grams (range 0.22 - 0.24) were tagged with half-length coded-wire tags in a production tagging operation at the Snettisham State Fish Hatchery, Juneau, Alaska. Fish were tagged between mid-May and mid-June, 1987 and sampled for histologic examination between late-May and July 1, 1987. Chum salmon collected for histologic examination were sampled on either the 7th or the 15th day post tagging and coho salmon were sampled on either the 19th or the 21st day post tagging. All samples of fish were preserved in Bouin's solution, processed by standard histologic techniques, and embedded in paraffin. Tags were carefully dissected from paraffin-embedded snouts with microdissecting tools under a dissecting microscope. Tag placement was noted and a sketch made. After the tags had been carefully removed, 5 µm thick sections were cut and stained with hematoxylin and eosin (H & E), and examined microscopically.

RESULTS

Chum salmon

Damage to the mainstem olfactory nerves was identified in 18% of the fish examined (36 of 200). Damage varied from substantial degeneration of the mainstem olfactory nerve to relatively

inconspicuous cytoplasmic swelling in nerve fascicles and individual nerve cells. In all cases, observed nerve damage corresponded directly with tag placement; i.e. if the left nerve was damaged, the tag was recovered from the left side of the head.

Coho salmon

Nervous tissue damage was observed in 37% of the fish examined (55 of 150). In the majority of the affected fish (35), the tags had been placed into the frontal lobe of the brain. While this deep placement undoubtedly increases tag retention, the brain should not be considered a desirable tag placement area.

DISCUSSION

The mainstem olfactory nerve damage observed in both the chum and coho salmon, and the questionable tag placement (frontal-lobe of the brain) in the coho salmon is a primary concern. Nervous tissue has a low regenerative capacity and tag placement into nervous tissue is likely repair completely, if at all. Olfaction would be affected, relative to the extent of olfactory nerve damage. It is probable that any impact on behavior would also be dependent on the degree of nervous tissue damage.

Traditional quality control techniques for assessing tag placement in salmonid fish involves simple dissection and gross examination, which identifies the general area where tags have been placed. This appears satisfactory when fingerling or yearling fish, are tagged (personal observation, J. Morrison), but when fry, weighing less than 1 or 2 grams, are coded-wire tagged, histological examination appears essential in assessing actual tag placement and for detecting any induced tissue damage. Nervous tissue damage is a potential bias that has not been considered in past studies. The above observations suggest the tagging process, itself, should be a consideration in studies involving coded-wire tagged fry.

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**PORTABLE PLANTING & FEEDING
EQUIPMENT MODIFICATIONS**

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Arcata, CA 95521 (707) 822-0592**

The necessity to better utilize trucks and equipment on hand led to the fabrication of steel framed racks and other modifications to planting and feed equipment allowing us multiple use of the vehicles.

quick removal and multiple use of our trucks. A description of the construction and a material list follows.

Summary of methods:

1. The oxygen bottle transfer rack was fabricated to insure safe, strain-free removal and replacement of 251 cu. ft. cylinders. A material list and illustration of the construction of such a labor saving device is included on the following pages.
2. The construction of portable frames for our 400 gallon and 1400 gallon tanks allowed us

Results and advantages:

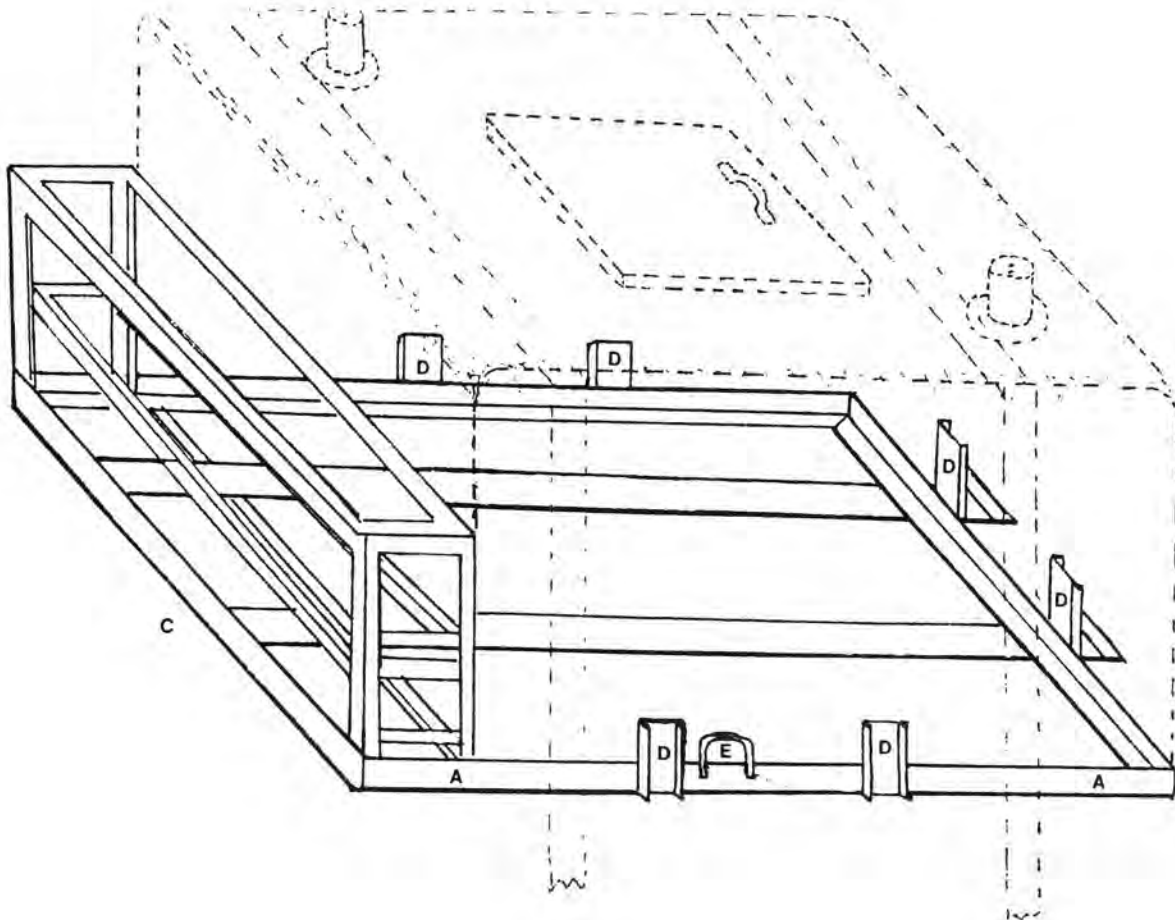
1. Feed truck is now used on long trips allowing highway mileage and a better running vehicle.
2. The O₂ bottle transfer rack using a fork lift insures a safe method of handling heavy bottles.
3. The fabrication of a steel frame for planting tanks and oxygen bottles insures easy removal using our fork lift with extensions.

PORTABLE 400 GALLON TANK & O₂ BOTTLE RACK

- A) Steel framing 3" angle iron x 91" long x 47" wide (drilled 1/2" holes for securing to 1 ton truck chassis).
- B) 1/2" round steel rod- for fork lift removal welded 8" x 3".
- C) O₂ bottle rack for two cylinders:
 - 1) 27" tall and 2" angle stock.
 - 2) 9 1/2" wide x 9 1/2" tall inside dimensions (2 compartments).
 - 3) 1" flat iron cage door to protect gauges, valves, and contain bottles.
 - 4) 4 each 2" x 2" angle iron x 46" long slide-in rails for O₂ bottles.
- D) Side and rear supports to contain fiberglass tank- 4 each 2" x 12" channel iron welded to frame.

Also needed:

2 each 14,000 lb. test cargo straps- secured to chassis frame with steel hooks and ratchet welded to frame under truck.



OXYGEN BOTTLE TRANSFER RACK USING FORK LIFT

Materials and Dimensions (Mild steel)

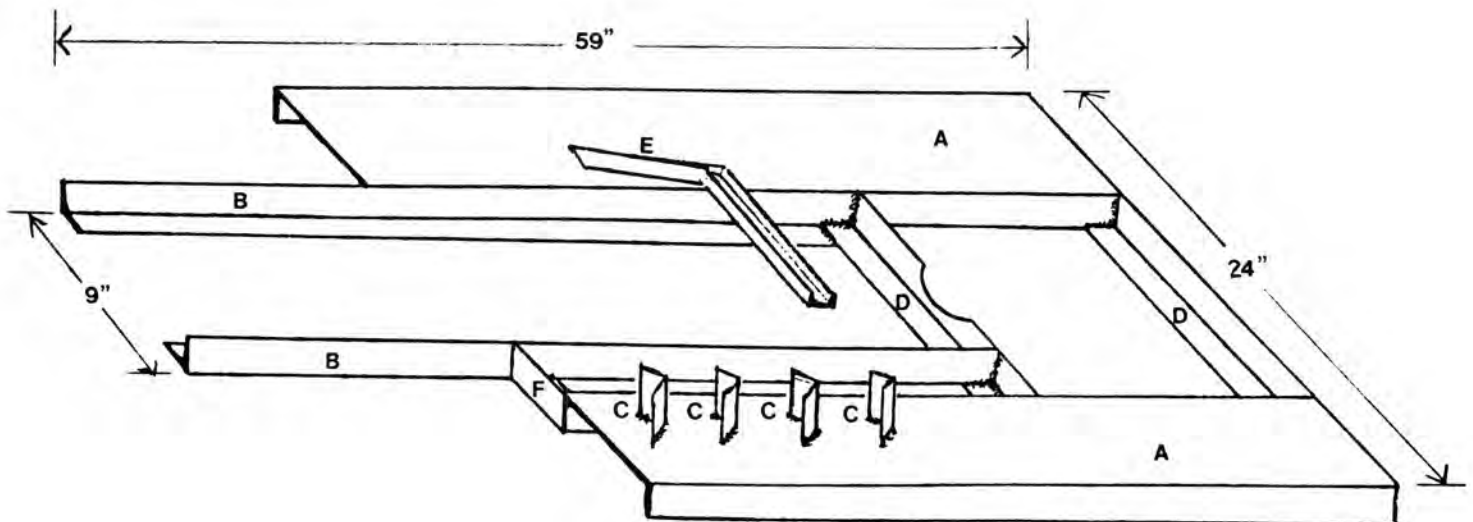
- A) 2 each fork lift channels 6" x 2" x 39".
- B) 2 each angle iron bottle supports 2" x 2" x 46" welded to channels.
- C) 6 each levering angle uprights 1" x 1" x 3".
- D) 2 each angle iron spreaders 2" x 2" x 12" (one cut out for bottle neck).
- E) 1 each channel iron top brace welded at an angle to prevent bottle from tipping out of bracket 1" x 2" x 8".
- F) 2 each small angle spacers to gauge bottle width 2" x 2" x 2" (welded to inside edge of fork channels).

Also needed:

1 piece 3/8" polypropylene rope with hook attached for pulling O₂ bottle from truck to lift rack.

1 each pulley block w/ 3" x 8" clevice attached to an anchor hook- cut from flat iron approximately 9" x 6" x 1/2" steel- hooks to fork lift frame.

1 each square aluminum tubing 1" x 36" for levering bottle into rack.



**CALIFORNIA DEPARTMENT OF FISH AND GAME
REARING AND HAULING DENSITIES**

**Ronald Ducey
California Department of Fish and Game
Nimbus Hatchery
2001 Nimbus Rd.
Rancho Cordova, CA 95670**

The data that follows was gather for no particular reason. Our hatchery coordinator, Ken Hashagen, and I were driving back from the conference last year and were wondering what our hatchery systems rearing and hauling densities are and how they compare to other fisheries agencies.

Rearing

I'll go through these pretty fast and point out some differences in comparison to other agencies but we're not as out of line as we thought:

1. B.C. is rearing more fingerling per ft³ of water than California.
2. B.C. is also rearing more brood fish on the average that California.
3. Hot Creek Hatchery has the highest density on brood fish and is also having problems with them.

Hauling

Hauling densities on trout in California are quite high. Maybe too high.

1. In general, except for one truck, as tank size increases, lbs/ft³ of water decreases.
2. When we compared California fingerling average hauling densities to information supplied by B.C. fisheries, there was not much difference but is the California density of 11.33 lbs/ft³ of water too high?
3. When we compare California chinook fingerling average hauling densities to information supplied by Washington Department of

Fisheries, we find a big difference, 2.5 to 5.6 lbs/ft³ of water.

I believe a lot more work needs to be done on rearing and hauling densities. We don't want to under produce at our hatcheries or always be on the ragged edge of disaster. We also shouldn't raise a quality product and stress it during hauling to the point only a small percentage survive.

The following is some of the information I collected:

Rearing Densities

CDF&G Catchables- 6/lb - 2/lb:

- Highest Density- 5.0 lbs/ft³ (Fish Springs)
- Lowest Density- .21 lbs/ft³ (Mojave River)
- State Average- 2.51 lbs/ft³

CDF&G Trout Fingerlings- 16.1/lb or smaller:

- Highest Density- 2.39 lbs/ft³ (Fish Springs)
- Lowest Density- .08 lbs/ft³ (Mojave River)
- State Average- 1.10 lbs/ft³

B.C. Fisheries Branch Trout Fingerlings 16.1/lb or smaller:

- 2.2 lbs/ft³ (raceways, tanks, and troughs)

CDF&G Brood Fish:

Highest Density- 4.28 lbs/ft³ (Hot Creek 2 year olds)

- Lowest Density- .97 lbs/ft³(Mt. Shasta BN-U)

- State Average- .242 lbs/ft³

B.C. Fisheries Branch Brood Fish:

- 3.0 lbs/ft³(raceways)

CDF&G Chinook Salmon Fingerlings 60.1/lb or smaller:

- Highest Density- .54 lbs/ft³(Nimbus)

- Lowest Density- .07 lbs/ft³(Mad River)

- State Average- .30 lb/ft³

CDF&G Chinook Salmon Advanced Fingerlings 60/lb - 20.1/lb:

- Highest Density- 1.66 lbs/ft³(Trinity River)

- Lowest Density- .9 lbs/ft³(Mokelumne River)

- State Average- 1.13 lb/ft³

CDF&G Chinook Salmon Yearlings 20/lb or Larger:

- Highest Density- 1.33 lbs/ft³(Trinity River)

- Lowest Density- 1.33 lbs/ft³(Trinity River)

- State Average- 1.33 lb/ft³

WDF Chinook Salmon- All Sizes:

- .3 lbs/ft³(ponds, 10'x100'x4' trough, 1/2 acre ponds)

Truck Loading and Hauling Densities

CDF&G Trout Catachables 6-2/lb:

150 Gallon Tank-

- Highest Density- 34.6 lbs/ft³ (San Joaquin)

- Lowest Density- 14.9 lbs/ft³ (Fish Springs)

- State Average- 25.6 lbs/ft³

220-250 Gallon Tank-

- Highest Density- 35.2 lb/ft³ (San Joaquin)

- Lowest Density- 18.1 lbs/ft³ (Mojave River)

- State Average- 25.7 lbs/ft³

400 Gallon Tank-

- Highest Sensity- 37.4 lb/ft³(Fillmore)

- Lowest Density- 19.7 lbs/ft³(Silverado & Hot Creek)

- State Average- 28.6 lbs/ft³

600 Gallon Tank-

- Highest Sensity- 26.7 lb/ft³(Silverado)

- Lowest Density- 13.26 lbs/ft³ (Mt. Shasta)

- State Average- 18.19 lbs/ft³

1200 Gallon Tank-

- Highest Sensity- 20.8 lb/ft³ (Hot Creek)

- Lowest Density- 13.9 lbs/ft³ (Fish Springs)

- State Average- 16.5 lbs/ft³

1500 Gallon Tank-

- 24.22 lbs/ft³(American River)

2500 Gallon Tank-

- Highest Sensity- 18.7 lb/ft³ (Mojave River)

- Lowest Density- 10.5 lbs/ft³ (Crystal Lake)

- State Average- 14.67 lbs/ft³

2800 Gallon Tank-

- Highest Density- 19.5 lbs/ft³ (San Joaquin)
- Lowest Density- 15.0 lbs/ft³ (San Joaquin)
- State Average- 17.2 lbs/ft³

CDF&G Trout Fingerlings 16.1/lb or Smaller:

600-1200 Gallon Tanks-

- Highest Density- 11.33 lbs/ft³ (Mt. Shasta)
- Lowest Density- 4.36 lbs/ft³ (Fish Springs)
- State Average- 6.89 lbs/ft³

B.C. Fisheries Branch Trout Fingerlings 16.1/lb or Smaller:

600-1200 Gallon Tanks-

- 5.8 lbs/ft³(12 hours, O₂ only, no agitation or recirculation)

CDF&G Chinook Fingerlings 60.1 /lb or Smaller:

600 Gallon Tank-

- 2.5 lbs/ft³(Nimbus 1/2 hour)

WDF Chinook Fingerlings 60.1/lb or Smaller:

1000-2500 Gallon Tanks-

- 5.6 lbs/ft³(Up to 4 hours)

CDF&G Advanced Chinook Fingerlings 60-20.1/lb:

1200-2800 Gallon Tanks-

- Highest Density- 13.75 lbs/ft³ (Mokelumne)
- Lowest Density- 4.2 lbs/ft³(Nimbus)
- State Average- 8.45 lbs/ft³(1 1/2 - 2 hours)

WDF Advanced Fingerlings 60-20.1/lb:

1000-2500 Gallon Tanks-

- 7.5 lbs/ft³(4 hours)

CDF&G Yearling Chinook 20/lb or Larger:

- No trucking data available

WDF Chinook Yearlings 20/lb or Larger:

1000-2500 Gallon Tanks-

- 9 lbs/ft³

FISH TRANSPORT TECHNIQUES AND TRIBULATIONS

Steve Arnold
British Columbia Ministry of Environment
Fisheries Branch
Fraser Valley Trout Hatchery
Abbotsford, B.C.

INTRODUCTION

The British Columbia Fish Culture Section has been stocking fish throughout British Columbia since the early 1900's. Over time, advancements have been made in fish transport techniques. However, it has only been in the last few years that we have seen major technological changes.

During transport, we have always used some method of supplying oxygen to the fish. The introduction of commercial fish farming to our coast, has made more advanced equipment available to us.

In the last few years, staff at Fraser Valley Trout Hatchery, have been testing and modifying oxygen monitoring and supply systems. We were trying to develop a system, that would enable us to provide an ideal concentration of oxygen, at a low consumption rate.

OXYGEN RACK SYSTEM

The system we previously used, employed a tygon tubing rack. The oxygen rack consisted of a length of tubing that had been perforated with small holes, to allow oxygen to escape into the tank. The oxygen was then supplied by a 222 litre oxygen bottle connected to a pressure gauge. A supply line was then fed from the pressure gauge, through a manifold located inside the truck cab. The manifold contained three pressure gauges, which regulated the amount of oxygen being supplied to each tank. We found the pressure gauges to be limited in their accuracy. It was therefore difficult to maintain a constant volume controlled flow of oxygen to each tank.

DIFFUSER OXYGEN SUPPLY SYSTEM

The redesigned oxygen delivery system consists of three oxygen flow gauges. These flow gauges were mounted onto the

manifold, replacing the three pressure gauges. The tygon oxygen rack has been replaced by two 24 inch long ceramic micropore diffusers.

The flow gauges increased accuracy, at the same time eliminating the problem of wasting of oxygen. The diffusers supplied a very fine localized oxygen pattern. The localized oxygen pattern raised concern regarding dead spots within the tank. These dead spots were a result of the small surface area of the diffusers; it was very important to create a stable supply of oxygen throughout the tank.

AGITATOR CIRCULATION SYSTEM

To correct the problem of insufficient oxygen circulation within the tanks, a battery powered agitator was mounted onto each tank. Each agitator requires only 4 amps to produce 3200 rpm. The unit is specially designed to inject air into the water and disperse it via a high impact plastic propeller.

Follow up tests of the agitator circulation system indicated that levels of oxygen within the tanks were more consistent. General observation of the fish, during transport, showed that they travelled better; losses and viable stress levels were also lower.

OXYGEN MONITORING SYSTEM

A system for continually monitoring oxygen levels was needed. Tests using hand held oxygen meters, showed levels as high as 25 ppm. Experiments conducted at Fraser Valley Trout Hatchery, determined optimum transport levels of oxygen, to be between 14 to 16 ppm.

The oxyguard system measures dissolved oxygen. Each transport tank has a probe, which relays this information to a LCD readout, located inside the truck cab. The probe is a galvanic measuring element,

which produces a millivolt output, proportional to the oxygen present in the water. Oxygen diffuses through the membrane onto the cathode, where a chemical reaction creates a electrical current. This current then flows, to a transmitter box, where the high and low oxygen alarms are located. The high and low alarms are set on the transmitter box; these settings range from 0 to 20 ppm. An increase or decrease in the oxygen levels from the preset readings will cause the alarm to sound. The alarm then alerts us to the need to correct the litre flow of oxygen.

SUMMARY

The oxyguard and circulation systems have eliminated many transport problems. The agitators have given us a better mixing of oxygen within the water column; they have also aided with the dispersement of other gases. The oxyguard system has enabled us to closely control the oxygen environment, during transportation.

The overall results of all this testing has been to reduce oxygen consumption by about 600% and to give fish culturists excellent control over the environment in which their fish are transported.

ADMINISTRATION OF ERYTHROMYCIN TO ADULT AND JUVENILE SALMONIDS WHAT ARE WE DOING AND WHAT DO WE KNOW?

Moderator and Organizer: Christine M. Moffitt
Department of Fish and Wildlife Resources
University of Idaho
Moscow, ID 83843

INTRODUCTION

Erythromycin has been used experimentally for 30 years in fish culture, yet we are still not sure how best to use it, and the use of the drug has not been approved by the US FDA or its counterpart in Canada.

At the University of Idaho, researchers have explored the usefulness of erythromycin as an injectable and oral drug for many years, beginning with studies of injection of adult chinook salmon conducted by Bill Klontz and his associates at Idaho hatcheries. In the early 1980's Ted Bjornn began studies that involved releases of coded wire tagged smolts to evaluate the long-term effectiveness of different strategies of oral administration of erythromycin to juvenile chinook salmon. In recent years, I have explored the palatability and the pharmacokinetics of oral administration of erythromycin. Erythromycin accumulates and is retained in the kidney of juvenile salmon up to 20 d following a daily oral administration of 0.1 g erythromycin per kg body weight for 21 d. Erythromycin may be particularly effective as a therapeutic because of this long retention time. In early 1989, Bonneville Power Administration contracted with us at the University of Idaho to

perform the tests necessary to register erythromycin as a therapeutic substance. The process of drug registration is defined carefully by FDA and protocols for all tests must conform to guidelines established for drug registration in minor species. To begin our studies we have been exploring the distribution and elimination of erythromycin administered to juvenile and adult salmon. In work conducted in our laboratory this past season, we found that erythromycin was retained for several days in the tissues of adult salmon following a single administration. We are still working on the analysis and modeling of pharmacokinetics of erythromycin in both adult and juvenile chinook. Then we will then conduct dose titration trials, toxicity tests and assess the environmental consequences of administration of erythromycin. As part of the background information needed for drug registration, we are compiling information on the form, dose, frequency of administration, efficacy, and any toxicity of past use of erythromycin to treat bacterial kidney disease in salmon. The members of this panel represent users in the state, federal and private aquaculture sectors. Each member of the panel will summarize of the experimental use of erythromycin in their respective facilities.

USE OF ERYTHROMYCIN AT FACILITIES OPERATED BY OREGON DEPARTMENT OF FISH AND WILDLIFE:

Craig Banner
Fish Pathology
Oregon Department of Fish and Wildlife
Oregon State University
Corvallis, OR 97331-3804

Erythromycin therapy for the control of bacterial kidney disease caused by *Renibacterium salmoninarum* appears to have

been quite successful at Oregon Department of Fish and Wildlife hatcheries.

Brood stock

Oregon Department of Fish and Wildlife has been inoculating brood salmon with erythromycin for almost 15 years. Therapy strategies vary among facilities, with fish receiving from 1 to 3 injections of erythromycin at a dose of 11 to 22 mg per kg body weight. Initially, erythromycin phosphate was used for brood fish inoculations, but more recently erythromycin base has been injected. The drug is delivered into the dorsal sinus by hypodermic needle and repeating syringe. Toxicity has been observed in some spring chinook stocks given injections at 22 mg per kg, however, losses have never been severe. Toxicity from erythromycin injection has never been observed in coho salmon.

Juvenile fish

Juvenile fish are fed erythromycin thiocyanate (poultry formula, Gallimycin 50-P, CEVA Laboratories) incorporated into the daily ration to deliver 4.7 g drug to each 45 kg of fish (approximately 0.1 g/kg). The poultry formula is incorporated into feed at 4 to 9 percent by weight, depending on the percentage body weight the fish are being fed. The medicated ration is administered first when fish are about 1 g in size. These fish are fed for a total of 21 d (14 d with a short break, then another 7 d). A second 21 d administration of medicated feed is given 40 to 60 d after completion of the first treatment. Toxicity has never been observed with erythromycin feedings, however, palatability has been a problem where the poultry formula has been incorporated into feed at greater than 7% by weight.

USE OF ERYTHROMYCIN TO CONTROL BACTERIAL KIDNEY DISEASE IN ADULT AND JUVENILE SALMON AT HATCHERIES OPERATED BY WASHINGTON DEPARTMENT OF FISHERIES

Mark DeCew, Fish Disease Laboratory,
Washington Department of Fisheries
P.O. Box 149 Salkum, WA 98582

BROOD STOCK

Erythromycin as Erythro 200 (CEVA Laboratories, 200 g/mL active erythromycin) is administered to adult salmon brood stock held at Washington Department of Fisheries salmon hatcheries. Although this drug is used primarily on spring chinook salmon brood stocks, a few stocks of coho and fall chinook adults receive erythromycin therapy. Injections are given subcutaneously into the dorsal sinus of each fish at a dosage of 20 mg erythromycin per kg of body weight. The purpose of the injection of erythromycin is threefold: 1) to control bacterial kidney disease (BKD) in the adult; 2) to reduce transmission of the causative agent of BKD, *Renibacterium salmoninarum*, from the adult to the ova; and 3) to provide therapeutic levels of erythromycin in the developing

eggs and fry. Adult spring chinook arrive at the hatcheries from April through July and receive from two to four injections during the holding period prior to spawning. The first administration is usually given on June 1 and subsequent injections are administered at 30 d intervals following the first. At some hatcheries, the final injection is given 10-14 d before the first spawning day, so therapeutic levels of erythromycin may be attained in the developing eggs and fry. Due to their short holding period, fall chinook and coho brood stock receive only one or two injections of Erythro 200. An untoward reaction has occurred in adult spring chinook salmon, where about 1% or less of the stock become jaundiced and die.

Stocks of adult spring chinook with a history of BKD have received erythromycin

for the past several years. In the years prior to this program, BKD incidence and mortality were high. Since the implementation of the injection procedure, loss and BKD incidence has been much lower. Two or more injections per season appear to reduce the level of infection. Studies using experimental lots of adults are currently being conducted to further evaluate efficacy of the drug.

JUVENILE FISH

Erythromycin thiocyanate (as Gallimycin-50p), is fed to spring chinook, fall chinook, and coho salmon to prevent and control BKD. Gallimycin-50p is incorporated into a moist fish feed pellet at 4.5 to 9.0% of the ration and fed for 21 d, providing a dosage of 99 mg erythromycin per kg of body weight. Starting in the 1988 season, treatments were administered before BKD epidemics had occurred. Most stocks of spring chinook were fed Gallimycin in early spring and again in mid-summer at a fish size of approximately 1.5 and 10 g, respectively. Other stocks were fed Gallimycin in the summer and then again in the fall, at fish size of approximately 10 and 25 g, respectively. When BKD epidemics have occurred during the spring or summer, therapeutic treatment with Gallimycin-50p has usually controlled mortality. However, when epidemics occurred in yearlings during the winter and spring, efficacy has been less apparent, and the treatments were associated with several obstacles. These fish were a fairly large size 40-70 g), and the water temperature

was low (40 F), which prevents the fish from consuming the usual therapeutic feeding rate of 1.5-2.0% body weight per day. Increasing the percentage of drug in the diet, from 4.5 to 9.0%, to compensate for the low feeding rate, seemed to cause feed refusal problems. An additional factor is that the cost of therapy increases as the fish attains a larger size.

Adverse drug reactions have occurred in spring chinook treated with Gallimycin-50p. During the treatment period and for 14 d post-treatment, spring chinook salmon may develop tetany and die, if subjected to the stress of handling. When all handling procedures are avoided, there are no untoward reactions. Some groups of yearling coho being fed Gallimycin-50p have become heavily infected with fungus and have suffered high mortality; however, it is known whether this condition was caused by the drug.

Evaluation of the efficacy of Gallimycin-50p, when the drug is fed at low water temperatures (40 F) to yearling chinook seems to be observed by the slow attrition of moribund fish from the population. Because BKD is a very slow developing disease, moribund fish may live throughout the treatment period. In some cases it has been observed that mortality will decline within about 10 d following the 21 d treatment period. Studies are planned to evaluate the efficacy of using Gallimycin-50p in relation to water temperature, fish size, and other factors.

USE OF ERYTHROMYCIN IN SALMON CULTURE FACILITIES OF IDAHO DEPARTMENT OF FISH AND GAME

Bill Hutchinson
Idaho Department of Fish and Game
600 South Walnut
Boise, ID 83707

Bacterial kidney disease is a major problem affecting chinook salmon in Idaho's state fish hatcheries. Losses in adults and

juveniles continue despite long standing treatments with erythromycin.

BROOD STOCK

Erythromycin has been used in treatment of adult chinook since mid-1970's. Both water soluble and oil or other base forms have administered into the dorsal sinus. Currently, Idaho state hatcheries are using erythromycin phosphate in an aqueous solution of 250 mg/L and a dosage of 10-11 mg/kg. Adult salmon are injected subcutaneously on the dorsal surface once at the time of collection.

Erythromycin injections of adult salmon are viewed as beneficial in reducing pre-spawning mortality. Although the Rapid River and Sawtooth hatcheries have seen improved survival rates in adults during those years of erythromycin injection, control groups of uninjected fish were not used, primarily because of the low numbers of adults available and the importance of each and every fish.

We have no real evidence of toxicity of erythromycin in adults. Jaundice and ulcerations at the injection site have been observed; but no conclusive results can be attributed directly to erythromycin injections.

JUVENILE FISH

Until 1988, juvenile chinook salmon had been fed erythromycin thiocyanate in the ration on a case-by-case basis, usually after mortalities resulting from BKD appeared. Treatments were administered by feeding Gallimycin 50 at 4.5 g (active ingredient) per 100 lbs of fish for a period of 21 d.

Currently Idaho Department of Fish and Game hatcheries are administering all juvenile chinook erythromycin in the ration. Fish are fed twice during the rearing cycle. The initial feeding begins during the spring when the fish are approximately 250-400/lb, and the second treatment is administered in early fall prior to a decrease in water temperature.

Promising results were experienced at Sawtooth Fish Hatchery during 1988. The group fed erythromycin exhibited a 0.9% mortality rate during the rearing cycle and had a 5% incidence of BKD (using FAT) at release, the control group had a 4.2% mortality rate during rearing, and an 19% incidence of BKD upon release. Needless to say, further testing is needed, but because of the importance of every fish, I doubt if extensive use of unfed control groups will be evaluated in Idaho hatcheries in the near future.

Up until 1989, adverse effects of fish fed erythromycin were not seen. This year several hatcheries experienced mortalities, during handling, after the second treatment of erythromycin. Although mortalities were not extensive, they were alarming. Affected fish exhibited tetany while being handled or stressed then died. Although this type of mortality was not consistent at all of Idaho's hatcheries, it was significant enough to cause us concern. Samples of fish were sent to the University of Idaho for evaluation. Results will be evaluated and may cause us to modify our erythromycin feeding regime next year.

USE OF ERYTHROMYCIN TO CONTROL BKD IN THE PRIVATE HATCHERIES

Jack Ganzhorn
Ore Aqua-Foods, Inc.
88700 Marcola Road
Springfield, OR 97478

BROOD STOCK

The injection of erythromycin in brood fish for control of BKD in the adults and/or in the progeny is widely practiced in private

aquaculture operations. Species injected include coho, chinook, rainbow trout (to control a Lactobacillus), and Atlantic salmon. Injected animals are not used for human or animal consumption.

Erythromycin (CEVA Erythro 100 or 200) is either injected from the bottle or mixed with oxytetracycline in a alcohol/acetic acid diluent. Generally 10-20 mg/kg fish are injected in the dorsal sinus (variable injection site). The dose is often governed by regulatory concerns, eg., Chile requires 11 mg/kg into adults before importing their eggs.

The frequency of injection depends on the reason for injection. For survival of brood fish, fish are normally injected upon arrival at holding facilities and possibly on a monthly basis until spawned (eg., spring chinook). Frequent injection protocols require extra care in handling. When the goal is to achieve a therapeutic dose in the eggs, injections are conducted within a few weeks or even a few days prior to spawning.

No controlled experiments were reported in the US; however, observations indicate that erythromycin injections do influence the incidence of BKD in the brood population. Some research is being conducted in BC, Canada (academic sector) and the primary focus is to search for more broad-spectrum drugs, especially for use in Atlantic salmon. No cases of toxicity have been reported.

JUVENILE FISH

Use of erythromycin in the ration is rare in the private sector due to its status as an

unregistered drug. Those companies that feed it under an INAD, have seen evidence that it is effective in controlling BKD, especially in saltwater. The uniqueness of each operation and the timing of treatments makes it difficult to generalize concerning its efficacy. Domsea, Anadromous, and OreAqua are the only private entities under an INAD permit for use of erythromycin in the US. Use in Canada is very rare.

Gallimycin 50 is incorporated into diet, and administered daily at 100-110 mg drug per kg body weight, generally for 21 d. Treatments are conducted at various times during the freshwater phase of smolt production or with farmed stocks in both freshwater and saltwater. Timing is contingent on observed incidence of *Renibacteria* by FAT analysis.

Controlled experiments have been conducted at Anadromous. Results indicated that erythromycin was effective in controlling BKD in saltwater; however, in freshwater, the disease process was only suppressed temporarily.

No cases of toxicity have been reported; however, significant problems with palatability were noted in British Columbia.

USE OF ERYTHROMYCIN IN FISH AND WILDLIFE SERVICE HATCHERIES

Jim Warren
U.S. Fish and Wildlife Service
911 NE 11th Ave.
Portland, OR 97232-4181

BROOD STOCK

Injection of erythromycin into the dorsal sinus of prespawning adult salmon is common in Fish and Wildlife Service hatcheries. Dosages of 11 mg erythromycin per kg body weight are most standard. In most cases, using the erythro 200 form of the drug may be better than using a larger

volume of erythro 100 (100 mg/mL active erythromycin) because less volume of carrier is injected to get the desired dosage. Most adult salmon are injected one or more times, usually at 30 d intervals, depending on how well the holding pond facilitates can segregate newly arriving fish from those arriving earlier. Jaundice toxicity has been observed at Warm Springs NFH

when water temperatures in the river rise above 15C (60F). Hatchery personnel found that during elevated river temperatures, injecting salmon at levels close to 22 mg/kg, using the Erythro 200 form of drug caused jaundice. When they switched to a lower dosage (11 mg/kg) and also switched to the erythro 100 form of the drug, they had fewer problems. Whether this toxicity is caused by the drug or carrier is not known.

The injection of adults, as they pass through the Warm Springs NFH weir, has increased redd counts in the Warm Springs river system. Pre-spawning mortalities are reduced but, little or no control of the tests have been maintained.

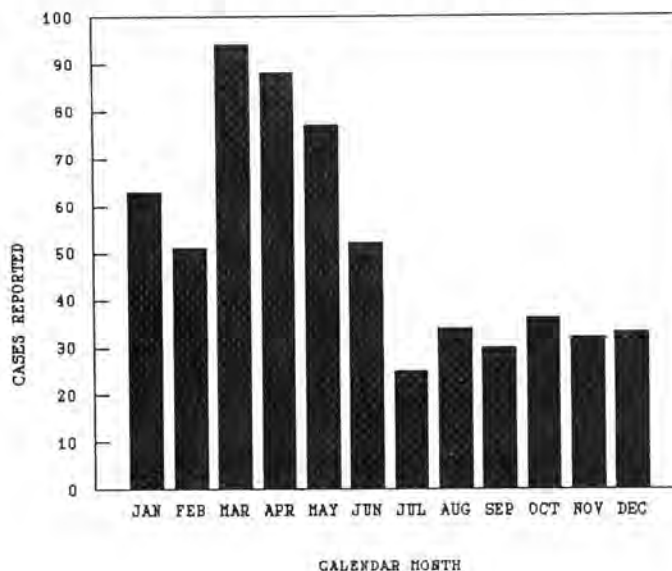
JUVENILE FISH

Usually erythromycin thiocyanate (as Gallimycin 50-P) is fed at the rate of 4.5 grams per 100 pounds of fish (100 mg/kg) for 21 d. We usually do not feed erythromycin prophylactically, but maybe we should in some situations. Therapeutic treatments are not given until the *Renibacterium salmoninarum* prevalence, in apparently healthy, random, mid-pond fish,

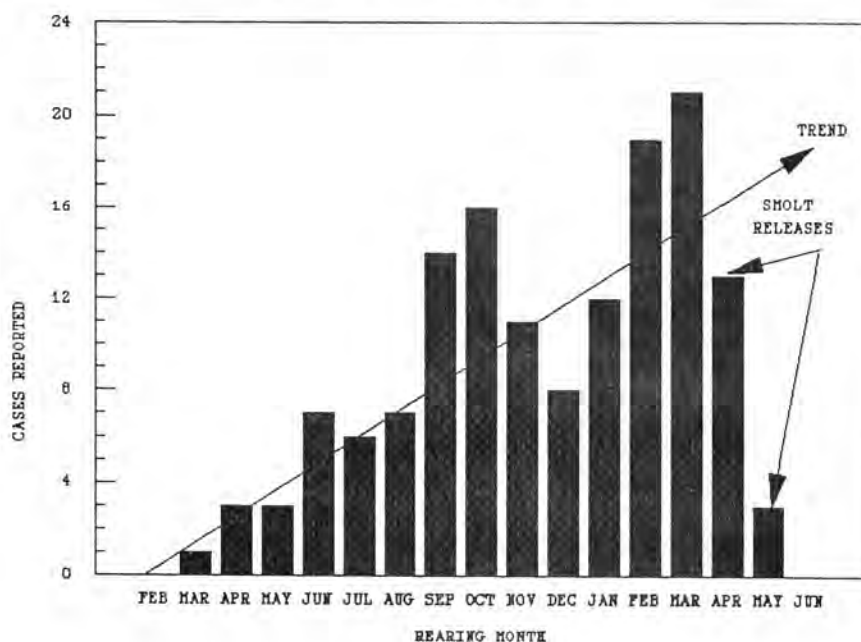
exceeds about 10 percent. This rule-of-thumb is not uniformly applied, but I feel that a prevalence $\geq 15\%$ is the level at which BKD itself will cause decreased contribution and hatchery returns. Adult injection appears to reduce the need to treat juveniles. In most cases the greatest increases in BKD is reported in spring chinook during the spring prior to release of smolts. Any effective therapy, especially in the 3 months prior to release could be expected to be beneficial. We have little controlled data on the actual efficacy of feeding erythromycin. John Cvitanich, through his qualitative FAT techniques has the best data I know of. He can assay tissues for the proportion of "bar forms" of *R. salmoninarum* which he interprets as being dead cells. I tend to agree with his views but the process is laborious.

In recent years we have detected increased "fragility" in juvenile spring chinook for a week or two after a 21 d erythromycin treatment. If fish are handled, tetany followed by mortality occurs in some fish. The disease signs are not dissimilar to a B-vitamin deficiency. If the fish are not handled for a couple of weeks, they show no further signs of distress that can be associated with the treatment.

SEASONAL DISTRIBUTION
BACT. KIDNEY DISEASE - ALL SPECIES



BKD IN JUVENILE SPRING CHINOOK
BY REARING MONTH



**INJECTION OF ERYTHROMYCIN TO CONTROL BACTERIAL KIDNEY DISEASE IN
BRITISH COLUMBIA**

Carl Wesby
Department of Fisheries and Oceans
Pacific Biological Station
Nanaimo, BC V9R 5K6

BROOD STOCK

The injection of brood stock with erythromycin has become a useful tool for the control of bacterial kidney disease (BKD). In our SEP hatcheries where the main problem is BKD, only erythromycin is injected, while in those hatcheries having prespawning losses due to mixed infections, a combination of oxytetracycline and erythromycin has been used effectively. The procedures described below are based on the results of experimental work carried out at the Capilano Hatchery in North Vancouver, B.C.

In 1982 the Fish Disease Control Program started experiments to determine whether prespawning losses in Capilano's early run

coho salmon, could be reduced or eliminated by the use of erythromycin injections. Early run coho salmon return to the hatchery in June but do not spawn until late October to mid-November. Erythromycin was injected into the dorsal sinus of 200 coho at a dosage of 20 mg/kg body weight and 200 control fish were injected with an equivalent volume of sterile saline. Three injections were given at approximately equal intervals over the 5 month holding period. Prevalence of the BKD (determined by FAT) at spawning was 0.6% in those injected with erythromycin and 21.7% in the saline controls.

However, prespawning losses remained high in the erythromycin injected group; therefore experiments in subsequent years

used a combination of erythromycin and oxytetracycline injections at a dose of 20 mg/kg of each antibiotic.

The following year, 300 fish were injected with the combined antibiotics and 300 controls were handled and anesthetized but not injected. In this experiment, to reduce handling stresses and to test the efficiency of a single injection of the combined antibiotics, the coho salmon received a single injection as they entered the hatchery in June. The results are shown below.

The third year, 500 early coho salmon received two injection of the erythromycin-oxytetracycline combination, the first immediately upon return to the Capilano Hatchery, and the second approximately 3 weeks prior to spawning. An equal number of control fish were handled and anesthetized but not injected. The group injected with antibiotic showed a reduction in the amount of external fungus and number of external lesions and a prespawning loss of only 12.6%. The uninjected control group suffered 67.4% prespawning loss. The injected fish yielded 89,000 viable eggs vs 6,300 for the control fish. The conclusions that can be drawn

from these results are: 1) erythromycin injection of brood stock can significantly reduce the presence of BKD; and 2) a combination of erythromycin and oxytetracycline injection can significantly reduce prespawning losses over long holding periods.

Another benefit of brood stock injection is the reduction of vertically transmitted BKD. A reduction of the number of bacteria in the ovarian fluid lessens the chance of vertical transmission. Research carried out by Dr. Trevor Evelyn at the Pacific Biological Station has shown that injection of brood stock 10-60 d prior to spawning can result in sustained bactericidal levels of erythromycin within the egg to further reduce vertical transmission of the disease.

We observed no toxicity problems during our experiments although occasionally some irritation was observed at the injection site on a few fish.

JUVENILE FISH

We have not experimented with feeding of erythromycin to juvenile salmonids.

| Parameter measured | Control uninjected fish | Injected with drug |
|---|-------------------------|--------------------|
| Incidence of <i>R. salmoninarum</i> (FAT) | 49.5% | 13% |
| Prespawning losses | 30% | 55% |

RELATIONSHIP OF OVULATION AND STRIPPING TIME TO EARLY SURVIVAL RATES IN RAINBOW TROUT (*Salmo gairdneri*)

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INTRODUCTION

The effects of egg retention in the body cavity of rainbow trout and determination of optimum stripping time post-ovulation have been examined by a variety of authors over a range of water temperatures (Springate, et al. 1984). Current spawning practice at Clear Springs Trout Company brood station located in Soda Springs, Idaho dictates sorting and spawning of a given lot of rainbow trout spawners at 9-12 day intervals, which exceed the maximum 10 day schedule recommended by Springate, et al. 1984. The objective of this study was to determine the optimum timing of fertilization post-ovulation as measured by early survival rates (eye-up and hatch) and develop a sorting/spawning schedule from the results.

MATERIALS AND METHODS

Five fish close to spawning were randomly selected from a group of two year old March spawning rainbow trout. Each fish was given an identifying fin clip, noted as fish #1 through #5, and separated from the rest of the lot. The fish were then checked daily for ripeness by anesthetizing each fish and applying abdominal pressure manually. The first day a fish expelled eggs was set as the date of ovulation (day 1). Approximately 250 eggs were removed each day after ovulation until the fish was completely spent. The eggs were stripped into a wire mesh colander, (allowing the natural ovarian fluid to drain) placed into 100mls of an artificial fertilization diluent and fertilized with 2.5mls of pooled and extended sperm. The sperm, diluent and eggs were gently stirred with a feather and allowed to rest fifteen minutes. The eggs were then rinsed with fresh water and allowed to water harden for one hour. After water-hardening was completed the eggs were placed into small

incubator cups made of one and one half inch PVC pipe sections cut to fit a Heath incubator tray, and wire window screen material glued to one opening. The cups were then placed into a Heath tray and incubated for twenty five days at 48 F (8.8 C). Daily treatments with a 1000.0ppm formalin drip for fifteen minutes and a 0.1 ppm titratable iodine fifteen minute drip were used to prevent fungal and bacterial infections. Live eyed eggs were counted at day 25 and percent survival was calculated for each day a fish was spawned. Live eyed eggs were returned to the incubator cups and allowed to hatch (day 45). The sac-fry were then counted and percent hatch was calculated for each day a fish was spawned. Data for each day post ovulation from all five fish were combined and average percent eye-up and hatch were calculated.

RESULTS

Although survival rates between individual females were highly variable, eyed survival rates were best from approximately 6-17 days post-ovulation (Fig. 1-5). Hatching survival rates showed similar trends through 13 days post-ovulation, followed by a subsequent decline in hatch rate regardless of eye-up percentage. Optimum combined eye-up survival rates occurred between days 6-19 post-ovulation, with hatch rate peaking from day 6-13 (Fig. 6).

DISCUSSION

The data suggest that, at the Soda Springs Brood Station with constant 48 F water, sorting and spawning can be scheduled in the range of nine to thirteen days to achieve best eye-up and hatch rates. Sorting intervals over thirteen days should be avoided. Four of the five females examined displayed a brief period of under-ripeness. Therefore, with ovulation set as day one a fish that expells only a

few eggs at sort should be returned to its original pond then spawned on the next scheduled sort.

The high inter-individual variation suggest factors other than post-ovulation timing may be important in early survival rates and indicate further needed research. account for lower then normal survival rates seen occasionally in egg lots.

ACKNOWLEDGEMENTS

We would like to thank Jim Parsons for his review and editing of this paper, also for preparing the graph slides.

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POST-OVULATION SURVIVAL FEMALE # 1

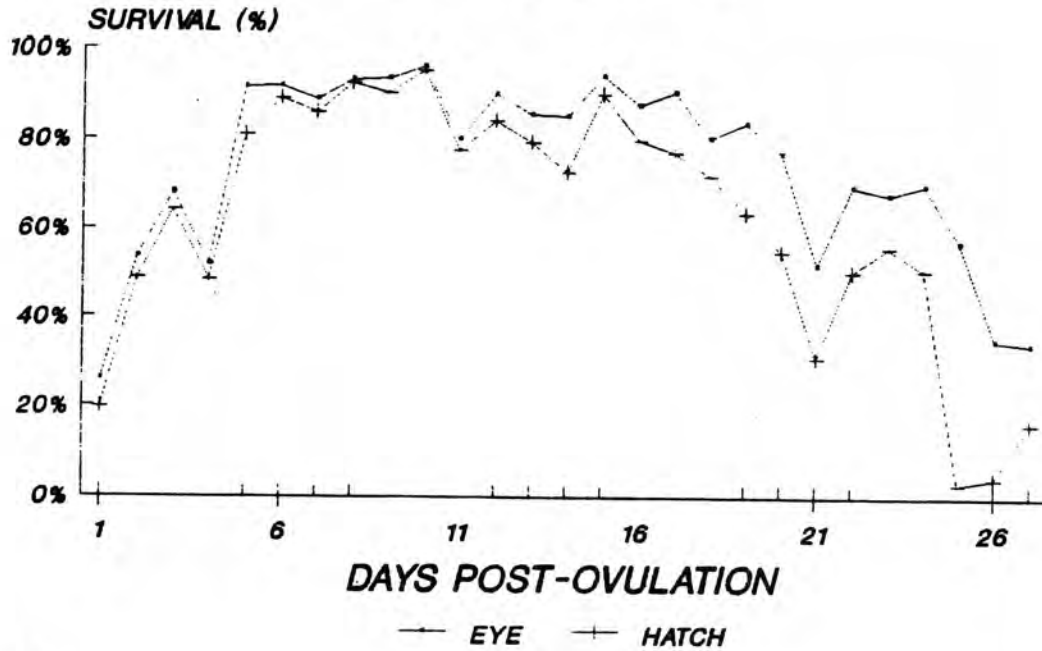


Figure 1.

POST-OVULATION SURVIVAL FEMALE # 2

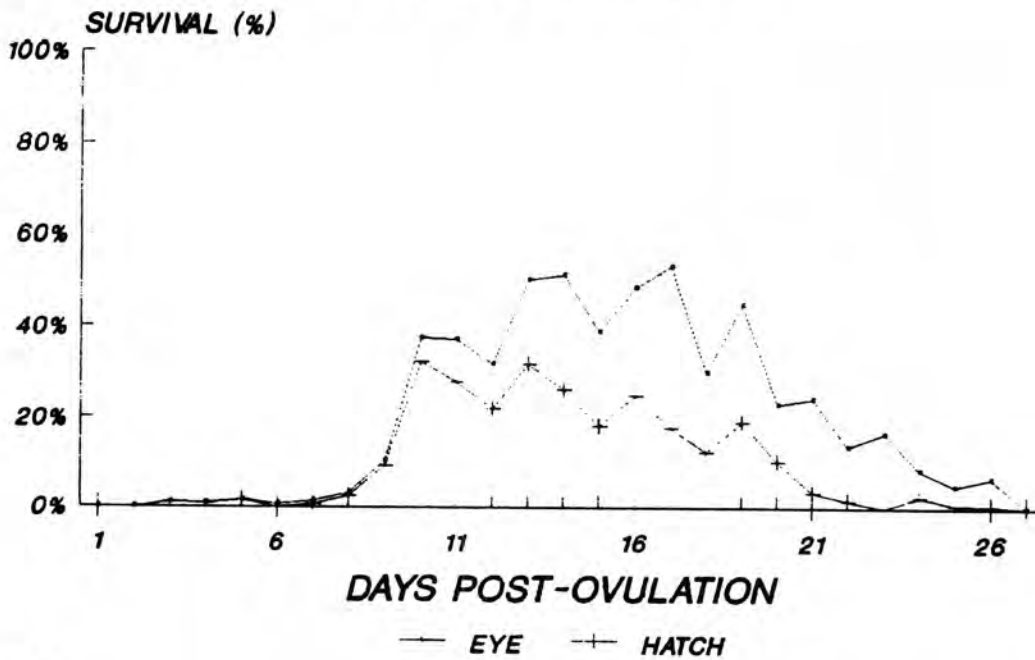


Figure 2.

POST-OVULATION SURVIVAL FEMALE # 3

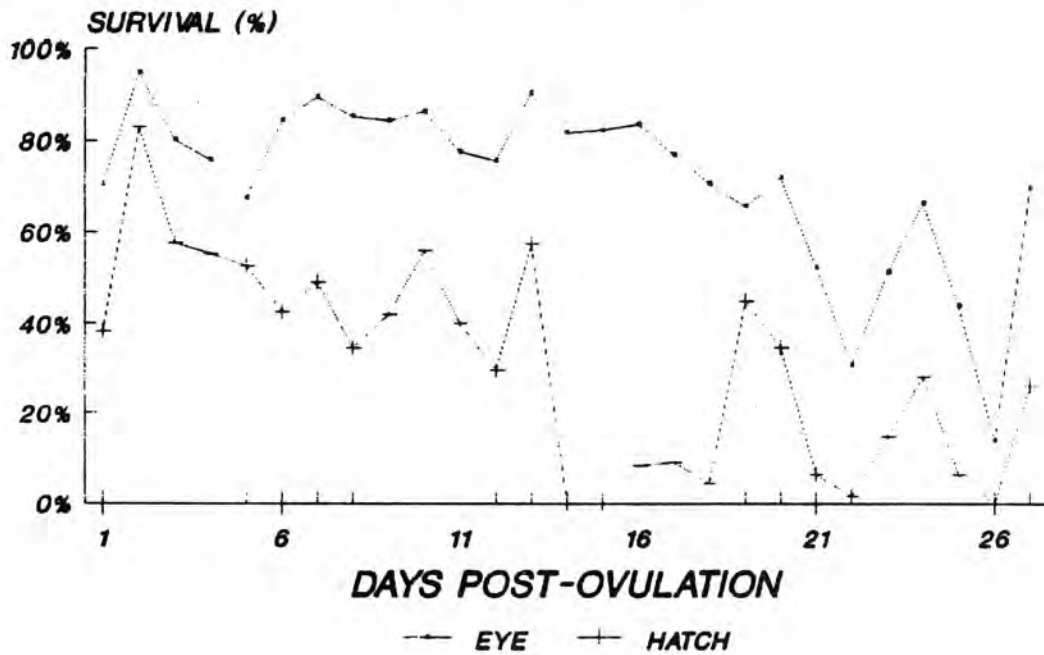


Figure 3.

POST-OVULATION SURVIVAL FEMALE # 4

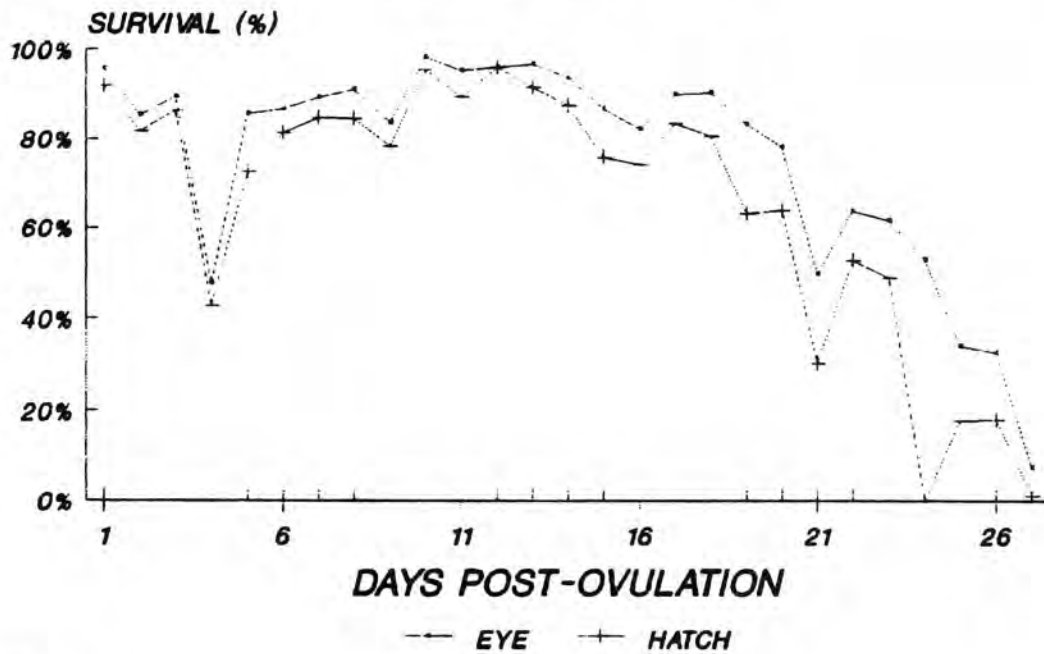


Figure 4.

POST-OVULATION SURVIVAL FEMALE # 5

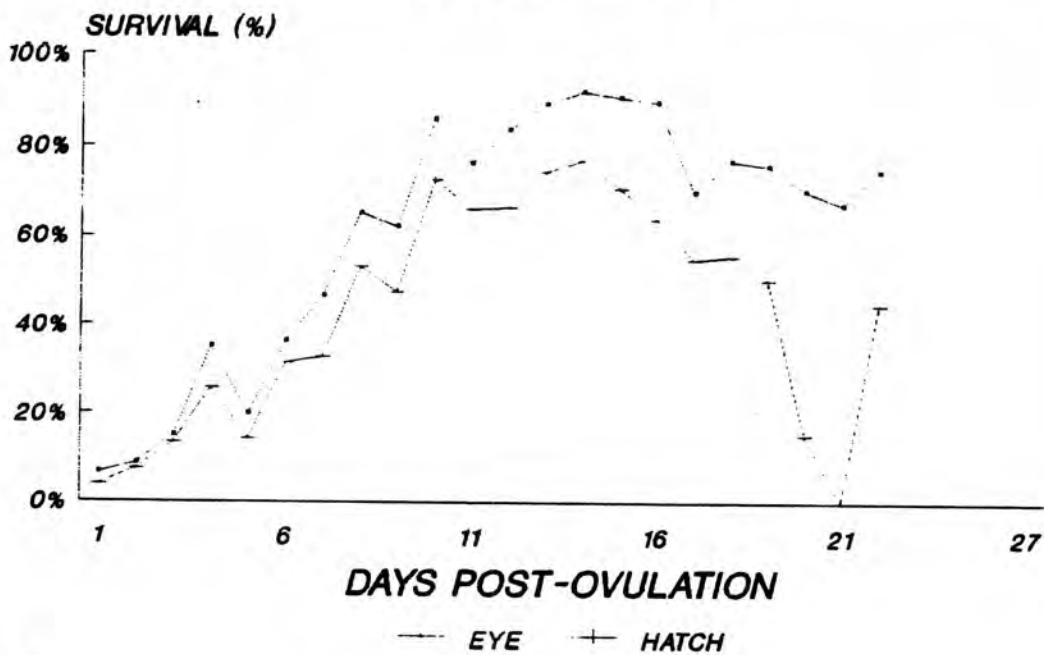


Figure 5.

POST-OVULATION SURVIVAL AVERAGE

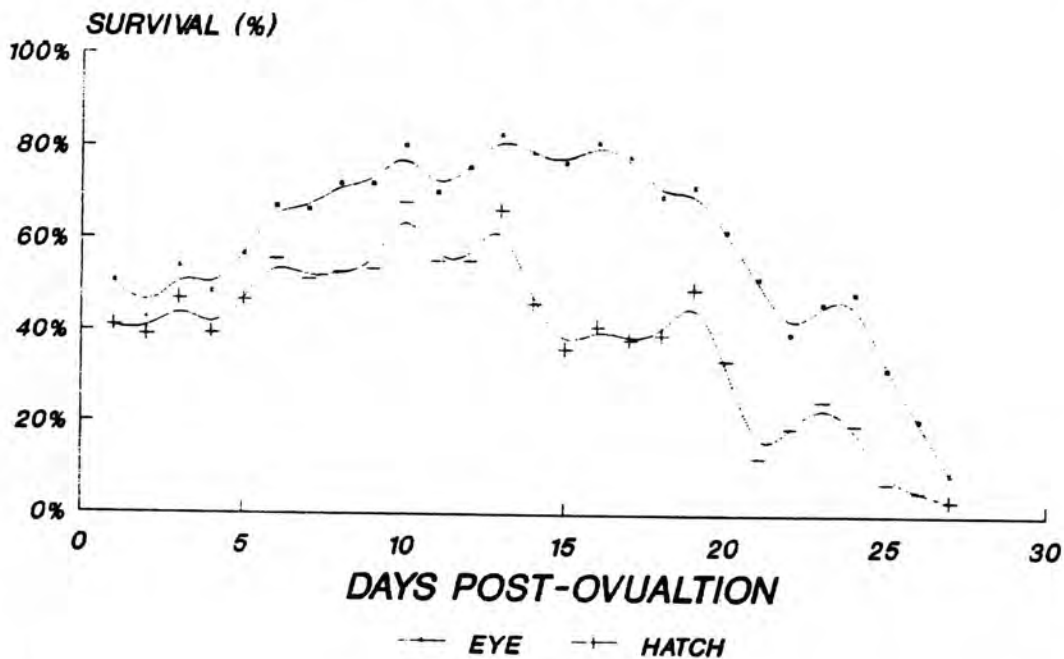


Figure 6.

THE EFFECT OF HANDLING AND/OR TRAUMA ON GREEN FERTILIZED EGGS RELATIVE TO TIME POST-FERTILIZATION

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The fact that developing salmonid embryos pass through a period of increased sensitivity during incubation has long been known to fish culturists. Mechanical shock resulting from egg handling procedures during this sensitive period may cause increased egg mortality. Rutter (1902) noted a "sensitive" time in developing chinook salmon (*Oncorhynchus tshawytscha*) embryos that lasted from day 6-16 after fertilization. Wales (1941) suggested that the sensitivity of steelhead trout (*Salmo gairdneri*) embryos gradually increased and then decrease throughout the developmental period. Davis (1956) and Leitritz and Lewis (1976) reported that trout and salmon eggs became progressively more sensitive during a period extending from 24-48 hours after water hardening until eye-up. Leitritz and Lewis (1976) also observed an extremely critical developmental period that occurred approximately midway to eye-up. Although none of these studies have specifically quantified degree of egg sensitivity or percent mortality over time, they have functioned to establish a basic philosophy amongst fish culturists that newly fertilized eggs are best left entirely alone until eye-up. At facilities that utilize vertical flow incubators, this philosophy commonly translates into a protocol which dictates that once eggs are put down in incubators, they are not even looked at until eye-up.

The present study was designed to determine at what stage of development (if any), and to what extent, salmonid eggs are sensitive to handling or mechanical shock. It was initiated based upon an identified need by the Great Lakes Lake Trout Restoration Program to be able to handle and transport either unfertilized eggs or newly fertilized green eggs. Currently, there is a considerable amount of similar interest with respect to a variety of other species. Fisheries

management programs are placing increased emphasis on rehabilitation or stocking programs involving wildstock or native fish populations. Virtually all of these programs, to at least some extent, will involve the transportation or handling of green fertilized eggs

MATERIALS AND METHODS

This study evaluated eggs collected from 6 strains (3 species) of trout: 1) lake trout (Marquette strain); 2) McConaughy rainbow; 3) Eagle Lake rainbow; 4) Yellowstone cutthroat; 5) Westslope cutthroat; and 6) Greenback cutthroat. Eggs were obtained from federal and state hatcheries in Wisconsin and Montana. Eggs were collected using either air spawning or standard hand stripping procedures. The number of females collected for each strain ranged from 9-47 (Ave=21). Eggs were fertilized with pooled samples of precollected milt. The number of males collected for each strain ranged from 4-47 (Ave=17). After fertilization procedures, all of the eggs from a given strain were pooled. Lake trout and Greenback cutthroat eggs were fertilized on station. Eggs from the other 4 strains were fertilized off station and transported to incubation facilities in a thermos-type jug containing a mixture of water and crushed ice. Transport times ranged from 1-2 hours.

Eggs from pooled samples were distributed randomly in small wire baskets and placed in vertical flow Heath incubators. Treatment groups ranged from 170-760 eggs/basket (Ave=438). Replicate treatments were conducted for each strain of rainbow and cutthroat trout. Only a single trial was conducted with lake trout.

Rainbow and cutthroat eggs were examined at 17 treatment times that

ranged from 6 hours to 20 days post-fertilization. Lake trout eggs were examined at 22 treatment times that ranged from 15 minutes to 24 days post-fertilization. All eggs were exposed to 2 treatment levels of mechanical shock at each treatment time; 1) "gentle shock", and 2) "hard shock". The gentle shock consisted of dewatering eggs and then dropping them from a height of 6 inches into 2 inches of water. This shock was intended to simulate the type of handling stress eggs might receive during "routine" hatchery manipulations (i.e. enumerating eggs, opening trays and picking eggs, early determinations of percent fertilization, etc). The hard shock consisted of dewatering eggs and then dropping them from a height of 6 inches onto a plastic surface (modified coffee can lid). The plastic was positioned at a 90 degree angle to the line of fall. This shock was intended to simulate the type of handling stress eggs might receive during shipping or transport procedures.

Lake trout eggs were incubated at approximately 43 F (11 T.U./day). Rainbow and cutthroat trout eggs were incubated at a constant 47 F (15 T.U./day). Egg viability was determined following shocking and hand picking of strongly eyed eggs.

RESULTS

The basic pattern of egg sensitivity with respect to time post-fertilization was very similar for all 6 strains evaluated. However, strain differences were observed with respect to degree of sensitivity to handling stress. In all strains eggs were more sensitive to hard shock treatment than gentle shock treatment. Results of individual strain evaluations are presented in Figures 1-6.

Lake trout eggs showed no sensitivity to gentle shock treatment through Day 6. Percent eye-up during this period was virtually the same as control eggs. On Days 8-14 gentle shock treatment resulted in an extremely slight decrease in eye-up (increased egg sensitivity). By Day 18, eye-up had returned to control levels. Although the effect of hard shock treatment of lake trout eggs resulted in an

almost identical pattern of egg sensitivity as observed with gentle shock treatments, egg sensitivity was greater. Hard shock treatment resulted in a slight decrease in eye-up on Days 1-6, a marked decrease (app. 40%) on Days 8-18, and had no effect on Day 23 (Figure 1). Lake trout eggs in this study eyed in about 45 days and hatched at 90 days.

McConaughy rainbow trout eggs showed even less sensitivity to gentle shock treatment than did lake trout eggs. With the exception of Day 12, gentle shock treatment resulted in no decrease in eye-up as compared to control eggs. On Day 12 however, gentle shock treatment did result in about a 20% decrease in eye-up. The effect of hard shock treatment on McConaughy eggs was dramatically different that its effect on lake trout eggs. Hard shock treatment resulted in an immediate, marked decrease in eye-up (app. 30%) at 6 hours post-fertilization. Egg sensitivity then increased progressively with time in an almost straight-line fashion with peak sensitivity on Days 10 and 12. Hard shock treatment on Days 10 and 12 resulted in an almost complete loss of egg viability (2.9 and 2.0% respectively). On Days 14, 16, and 18, there was a relative rapid, progressive decrease in egg sensitivity, and by Day 20, eye-up had returned to control levels (Figure 2). Day 20 eye-up had returned to control levels (Figure 2). McConaughy eggs in this study (as well as Eagle Lake rainbow and the 3 strains of cutthroat trout eggs) eyed in about 23 days and hatched in 40-43 days.

The effect of handling stress on Eagle Lake rainbow trout eggs was basically the same as with McConaughy eggs with respect to both treatment levels. Gentle shock treatment resulted in a very slight decrease in eye-up on Days 6-9, 30% decrease on Days 10 and 12, and had no effect at all other treatment times. Hard shock treatment resulted in an immediate, marked decrease in eye-up (app. 30%) at 6 hours post-fertilization. Although egg sensitivity held somewhat constant on Days 1-3, it then increased progressively and peaked on Days 10 and 12, at which time egg viability was virtually nil. Sensitivity progressively decreased on Days 14 and 16, and eye-up returned to control levels by Day 18 (Figure 3).

Yellowstone cutthroat trout egg sensitivity followed the same pattern with respect to both treatment levels as did the eggs of the two rainbow trout strains. Gentle shock treatment had no effect on egg viability with the exception of a slight decrease in eye-up on Days 10, 12, and 14. Hard shock treatment resulted in a progressive increase in sensitivity that peaked at Day 12, followed by a progressive decrease and return to control levels by Day 18. The major difference in the response of the Yellowstone cutthroat eggs to the hard shock treatment as compared to the rainbow eggs was that sensitivity increased much more gradually. Rather than an immediate, marked decrease in eye-up, egg sensitivity increased only slightly at 6 hours post-fertilization. It was not until Day 5 that it approached levels observed at 6 hours in the two strains of rainbow trout (Figure 4).

The pattern of Westslope cutthroat trout egg sensitivity was the same as observed for the two strains of rainbows and Yellowstone cutthroat trout. The only effect of gentle shock treatment was a decrease in eye-up on Days 9, 10, and 12. Hard shock treatment resulted in a progressive increase in sensitivity up to Day 12, a progressive decrease in sensitivity on Days 14 and 16, and a return to control levels of eye-up on Days 18 and 20. A major difference observed with Westslope cutthroat eggs was the extremely marked, immediate egg sensitivity to hard shock treatment. Hard shock at 6 hours post-fertilization decreased eye-up about 65% from that of control eggs (Figure 6).

Greenback cutthroat trout egg sensitivity followed the same basic pattern with respect to both treatment levels as did the other strains of cutthroat and rainbow trout. Gentle shock treatment indicated egg sensitivity only at Days 10 and 12. Sensitivity to hard shock treatment was most similar to Yellowstone cutthroat eggs. Greenback eggs showed less sensitivity to hard shock treatment up to Day 2 post-fertilization than the other strains evaluated (Figure 6).

In order to better determine both patterns and degree of egg sensitivity relative to the 3 species of trout evaluated, data from

the 2 strains of rainbow and 3 strains of cutthroat were combined (by species and with respect to treatment levels) and plotted along side of the lake trout data. This data was analyzed based on percent survival rather than percent eye-up (control group eye-up was considered to be 100% survival). This data is presented in Figures 7 and 8.

The pattern of egg sensitivity in response to gentle shock treatment was virtually identical in the rainbow and cutthroat trout. Egg sensitivity was first evidenced on Day 9 and lasted until Day 14. The pattern of egg sensitivity in lake trout was similar, but appeared to begin about 1 day earlier and last several days longer. The degree of egg sensitivity in response to gentle shock treatment was virtually identical in the rainbow and cutthroat, with peak sensitivity on Day 12 resulting in a 25-30% decrease in egg survival. The degree of lake trout egg sensitivity was considerably less than the eggs of rainbow and cutthroats, with peak sensitivity at Days 8-16 resulting in only a 7% decrease in egg survival. The pattern of egg sensitivity in response to hard shock treatment was virtually identical in the rainbow and cutthroat trout. Egg sensitivity began 6 hours post-fertilization, increased progressively up to Day 12, decreased progressively on Day 14, 16, and 18, and return to control levels by Day 20. The pattern of egg sensitivity in lake trout was basically similar although initial sensitivity increased more gradually and peak sensitivity was maintained for a longer period of time. The degree of egg sensitivity in response to hard shock treatment was virtually identical in rainbow and cutthroat trout. Percent survival decreased by 30% at 6 hours post-fertilization, by 95% on Day 12, and returned to control level by Day 20. Lake trout eggs were much less sensitive to hard shock treatment than rainbow or cutthroat eggs. During the period of peak sensitivity (Day 8-20), percent survival decreased only about 40% (vs 95% in rainbow and cutthroat).

DISCUSSION

The results of the present study support the basic findings of earlier investigations

that have examined the sensitivity of salmonid embryos to handling stress with respect to stage of development (Rutter, 1902; Wales, 1941; Davis 1956; and Leitritz and Lewis, 1976). In general terms, the sensitivity of salmonid eggs to mechanical shock appears to initially increase in a progressive, straight-line fashion up to a certain stage of development, and then decrease in a rapid, progressive manner. In rainbow and cutthroat trout, embryo sensitivity begins to increase relatively soon after fertilization, reaches a peak on Days 10 and 12 (150-180 T.U.), decreases rapidly, and appears to end by Day 18 or 20 shortly before eye-up.

Data obtained in this study showed that there was a considerable difference between the sensitivity of salmonid eggs to gentle shock and hard shock treatments. Whereas hard shock treatment resulted in an almost immediate decrease in embryo survival that progressed to a level where survival was virtually non-existent, gentle shock treatment had a relatively minor effect on embryo survival. The level of mechanical shock that developing embryos were exposed to by hard shock treatments functioned to provide an accurate description of true egg sensitivities. Embryo survival went from the level of control eggs to near zero, and then returned to control egg levels. The data leaves little doubt that salmonid embryos are indeed sensitive to handling during development, and that degree of sensitivity is relative to stage of development.

The fact that gentle shock treatments were apparently no severe enough to provide a clear picture of true egg sensitivity has some very interesting implications. The mechanical shock generated by gentle shock treatment (dewatering eggs and dropping them 6 inches into water) was intended to be representative of the degree of handling stress involved in "routine" hatchery egg handling procedures. If this is the case (and we believe it to be so), then based on the data from this study, there is no reason why newly fertilized eggs cannot be handled during a period of at least 5-7 days post-fertilization. In fact, with careful handling techniques, it is likely the egg manipulation can be conducted throughout the incubation period, with the

possible exception of a 3-4 day period of extreme egg sensitivity (150-200 T.U.). In any case, the concept of philosophy that newly fertilized eggs should be left completely alone until eye-up is not entirely sound, nor is it particularly conducive to maximizing hatchery production potential. At the very least, a culturist need not hesitate when considering pulling out a tray of eggs and attempting to make some sort of pre-evaluation of egg viability.

This study also indicated that there may be differences in the degree of egg sensitivity with respect to species. Lake trout eggs exhibited considerably less sensitivity to both gentle and hard shock treatment than did rainbow and cutthroat eggs. Gentle and hard shock treatments on lake trout eggs during the period of peak egg sensitivity resulted in a 7 to 40 % decrease in survival respectively. The same treatments on rainbow and cutthroat eggs resulted in a 25-30 and 95% decrease in survival, respectively. Based on this data, lake trout eggs appear to be more tolerant of handling stress than eggs of the other species tested. The effect of similar handling techniques on the eggs of other salmonid species remains to be established. Tolerance of lake trout eggs to handling is supported by data indicating that newly fertilized eggs can be transported in shipping containers (similar to the method normally used for eyed eggs) up to 72 hours post-fertilization with no loss of egg viability (Erdahl and McClain, unpublished).

SUMMARY

There is indeed a period of time during which salmonid embryos are sensitive to handling stress or mechanical shock. It appears the egg sensitivity in most species begins relatively soon after fertilization. Sensitivity continues to increase in a progressive manner, reaches peak levels, and then decreases to control egg levels prior to eye-up. The eggs of rainbow and cutthroat are most sensitive about 10-12 days post-fertilization (150-180 T.U.). Peak sensitivity in lake trout eggs occurs about 9-18 days post-fertilization (100-200 T.U.). Although the basic pattern of egg sensitivity is similar between species, lake

trout eggs are less sensitive to handling stress than are the eggs of rainbow and cutthroat trout. Based on the data for this study, it appears that the sensitivity of salmonid eggs to handling or mechanical shock during incubation is considerably less than is often assumed. With the possible exception of a period of 3-5 days, the eggs of most salmonids can probably be handled safely if proper caution is exercised.

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Figure 1. Effect of handling and/or trauma on green fertilized Lake Trout eggs relative to time post-fertilization

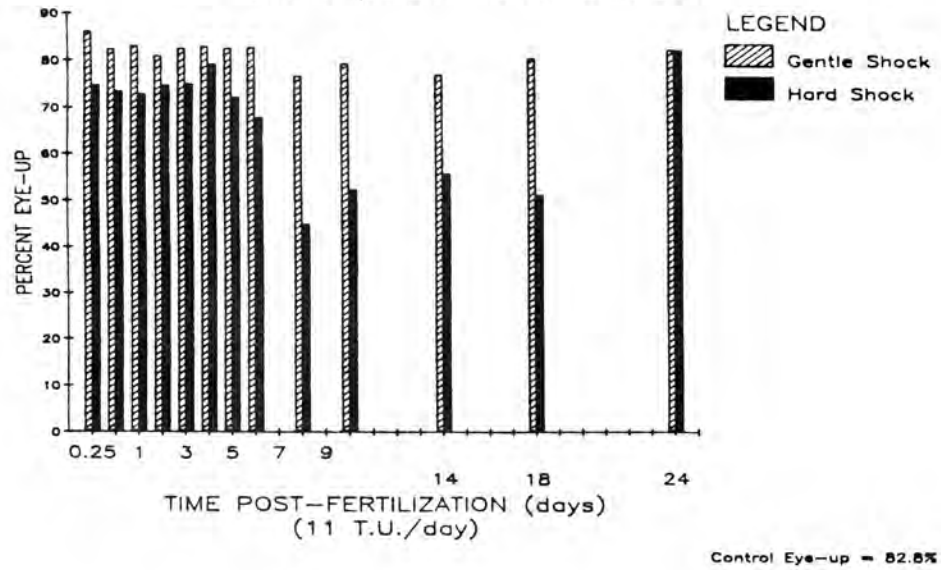


Figure 2. Effect of handling and/or trauma on green fertilized McConaughy RBT eggs relative to time post-fertilization

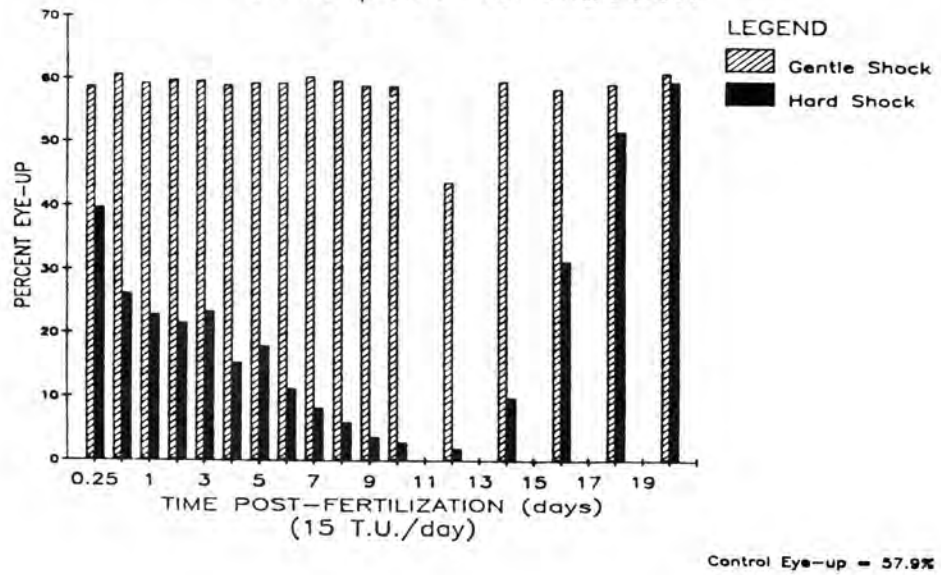


Figure 3.

Effect of handling and/or trauma on green fertilized Eagle Lake RBT eggs relative to time post-fertilization

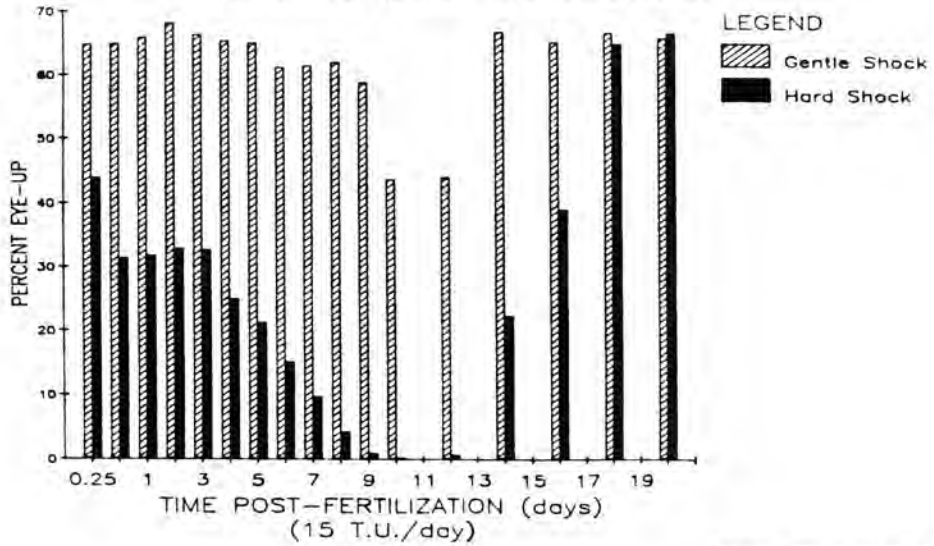


Figure 4.

Effect of handling and/or trauma on green fertilized Yellowstone CTT eggs relative to time post-fertilization

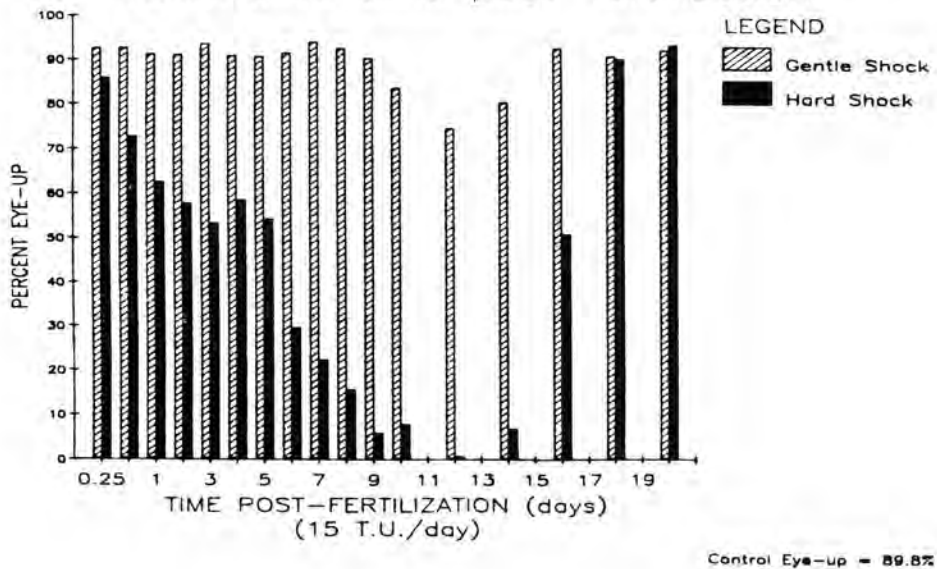


Figure 5. Effect of handling and/or trauma on green fertilized Westslope Ctt eggs relative to time post-fertilization

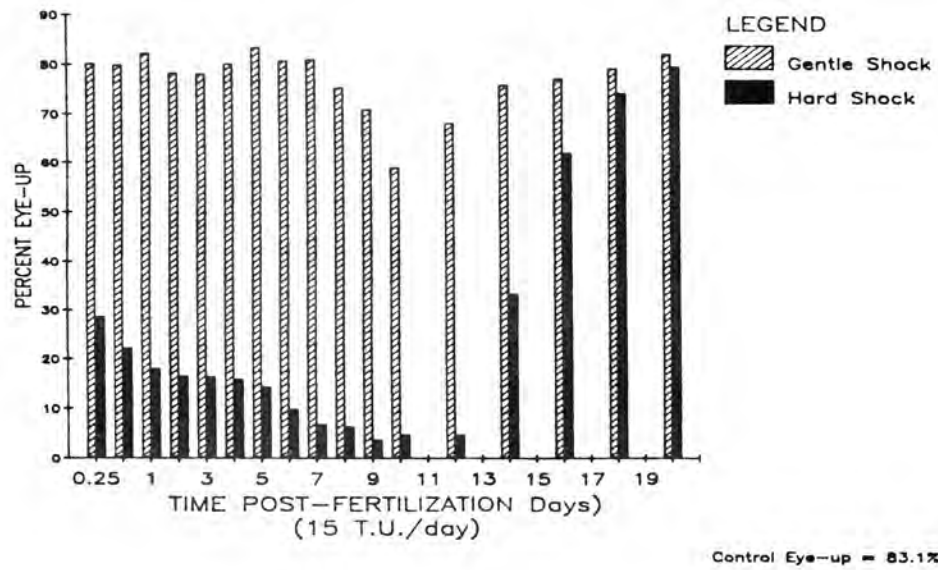


Figure 6. Effect of handling and/or trauma on green fertilized Greenback Ctt eggs relative to time post-fertilization

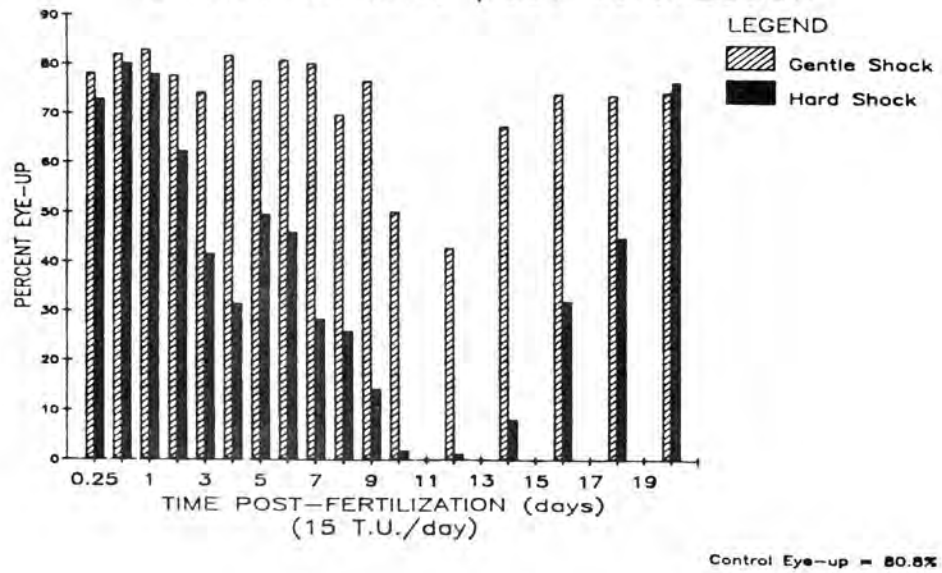


Figure 7. Effect of "Gentle Shock" on Green Fertilized Eggs Relative to Time Post-Fertilization

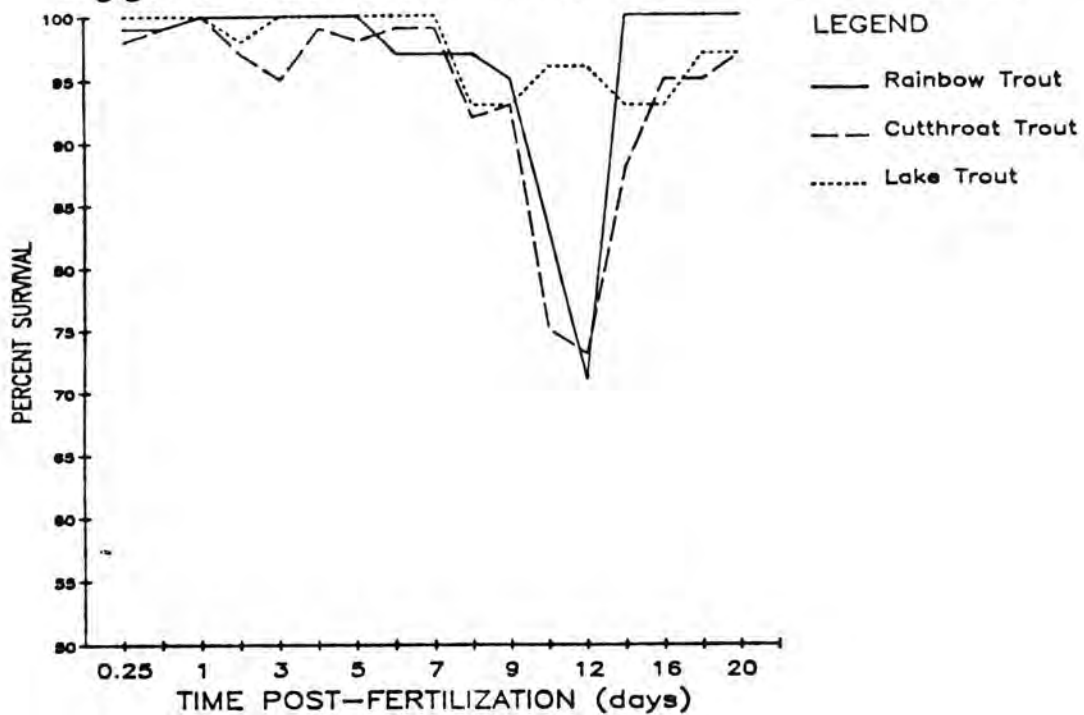
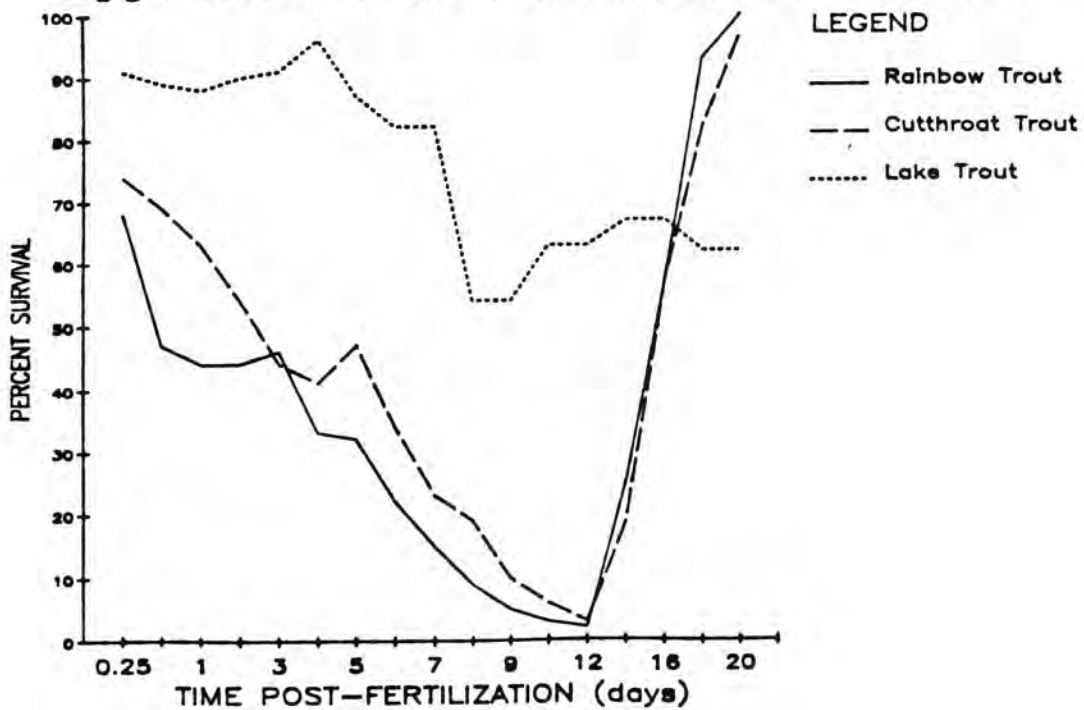


Figure 8. Effect of "Hard Shock" on Green Fertilized Eggs Relative to Time Post-Fertilization



COMBINATION OF EXTENDER SOLUTION AND IODOPHORE HARDENING
FOR DISEASE CONTROL AND IMPROVED SURVIVAL OF RAINBOW TROUT
(*ONCORHYNCHUS MYKISS*) EGGS

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ABSTRACT

The effect of several compounds used to improve the post-hatching survival of rainbow trout (*Oncorhynchus mykiss*) fry was investigated. The method for fertilization described by Hamor (1988) was followed up to the point of the first washing. For the first was, we used 2.25 mg/L ZnSO₄, 0.2 g/L NaCl, 0.1 g/L NaHCO₃, and 0.1 g/L vitamin C solution. The we immediately added 1:100 Argentyne. For the "second wash", we used 2.25 mg/l ZnSO₄, 0.2 g/L CaCl₂, 0.1 g/L vitamin C, and 0.1 g/L NaHCO₃. Subsequently, the eggs were hardened in a solution of 0.45 mg/L ZnSO₄, 0.04 g/L CaCl₂, 0.02 g/L NaCl, and 0.1 g/L vitamin C for one to two hours and then treated in a 1:300 Argentyne or Ovodyne solution before being placed in the incubator.

The addition of vitamin C in iodophore hardening resulted in a 5-10% increase of the number of fry over the same process without the vitamin. Use of iodophore compounds are recommended for control of the spreading of infectious diseases. A possible role of vitamin C in increasing fish resistance against IPNV and IHNV is discussed.

INTRODUCTION

Saline solutions to improve fertilization success were probably first described by Woynarovich (1955) and subsequently by many others (Erdahl et al. 1987, Hamor 1987, Wilcox et al. 1984).

In addition, survival seems to be influenced by introducing salts and other substances into the egg hardening solution (Hamor 1988); in particular vitamin C

seems to have the ability to increase survival (Hamor 1988, Felton and Halver 1988).

Eggs are also treated with disinfectants to prevent spread of pathogens like infectious pancreatic necrosis virus (IPNV) and infectious hematopoietic virus (IHNV).

The occurrence of IPNV in Alberta was first reported by Yamamoto (1974). In April of 1989 IPNV was again identified by our laboratory scientist, Bev Larson.

Ahne (1983) concluded that besides eggs and ovarian fluids, the seminal fluids of rainbow trout must be regarded as a source of IPNV contamination.

Egg fertilized with sperm exposed to IPNV did not show infection but the virus was subsequently isolated from fry. Dorson and Torchy (1985) detected IPNV in samples of ovarian fluid; however they did not find the virus in disinfected (50 ppm iodine) and washed ova.

Mulcahy and Pacho (1984) reported that more than 99% of IHNV, a vertically transmitted fish rhabdovirus was removed from suspension in less than one minute by adsorption to the surface membrane of sperm from two genera of salmonid fishes. The vertically transmitted IPNV was adsorbed to a lesser degree.

Ahne and Negele (1985) infected eggs of rainbow trout with IPNV. After transfer of the infected eggs into tanks with through-flowing water, the amount of virus associated with the eggs decreased with time and 24 hours after infection, the eggs were no longer infectious. The fry began to hatch 35 days after fertilization and the hatching rate was about the same

(95%) in both infected and control groups. Neither freshly hatched fry nor samples of fry taken were infected.

Yoshimizu et al. (1989) experimented on the survivability of IHNV in fertilized eggs of masu (*O. masou*) and (*O. keta*). They concluded that vertical transmission of IHNV is doubtful because the virus is apparently unable to survive in eggs before the eyed stage.

McAllister et al. (1987) detected IPNV in ovarian fluid of brook trout (*Salvelinus fontinalis*).

Methods for separating ovarian fluids and replacing them with saline solutions (Hamor 1969, 1987, Erdahl and Graham 1987, Erdahl et al. 1987, Wilcox et al. 1984) likely help to prevent the spread of diseases, in addition to increasing egg fertility.

Dorson and Torchy (1983) recommended fertilization in the presence of iodine as a means of preventing IPNV transmission, but they asked for further investigation on the efficacy of the method and to determine the toxicity of iodine to spermatozoa, especially as a function of pH. Castric (1985) described a method of applying 25 ppm iodine to eggs at the time of fertilization to treat for IPNV. However, Desautels and MacKelvie (1975) found that concentration of iodine necessary to destroy the IPNV should be at least 35 ppm for five minutes or more. The suggested treatment is potentially valuable but requires more investigation.

According to the U.S. Fish and Wildlife Service Proposed Fish Health Policy (1989), "The impact of IPNV on Atlantic and Pacific Salmon and lake trout appears slight, therefore eggs from IPNV positive lake trout or salmon populations (not steelhead) may be transferred to service facilities if all freshly spawned eggs are first water hardened in polyvinylpyrrolidone iodine disinfectant."

The effects of iodophore during egg hardening, in combination with rinsing and extender solution and supplements of vitamin C, were investigated to develop a

procedure to help prevent the spreading of fish diseases.

MATERIALS AND METHODS

Eggs and semen were collected originally from our second and third generation rainbow trout, Mount Lassen or Trout Lodge strains from California. Eggs were collected by strainers to separate them from ovarian fluid. After each female was stripped, the eggs were either rinsed in salt solution or, for controls, just transferred into a plastic or stainless steel pan. Semen was collected from three or four males to fertilize eggs collected from groups of five females.

After the introduction of semen, the eggs were continuously stirred and 0.5 L of an extender solution was added. Mixing of the eggs covered with the extender solution continued for about one minute. Following mixing, the eggs were left undisturbed for five minutes to complete the fertilization process, and then, with the use of a strainer, were placed in the first and then the second washing solutions for at least five minutes each. Finally, the eggs were treated with 1:100 iodophore in the first washing and with 1:300 iodophore for the second washing for five and ten minutes respectively, before they were placed on trays. The trays were divided into nine compartments, with about 1000 eggs in each compartment. Fertilization success was measured by calculating the percentage of fry from green eggs.

The fertilized eggs were split into two groups. One of the groups was subdivided into smaller lots and hardened in several different solutions using 10 and 20 minute time intervals.

The Graphics were enhanced following the procedure of Rudic (1956).

RESULTS

The different egg hardening treatments resulted in differences in fry survival (Table 1 and Figure 1).

TABLE 1.

Fry survival resulting from several concentrations of Argentyne solution used at two treatment times for egg hardening.

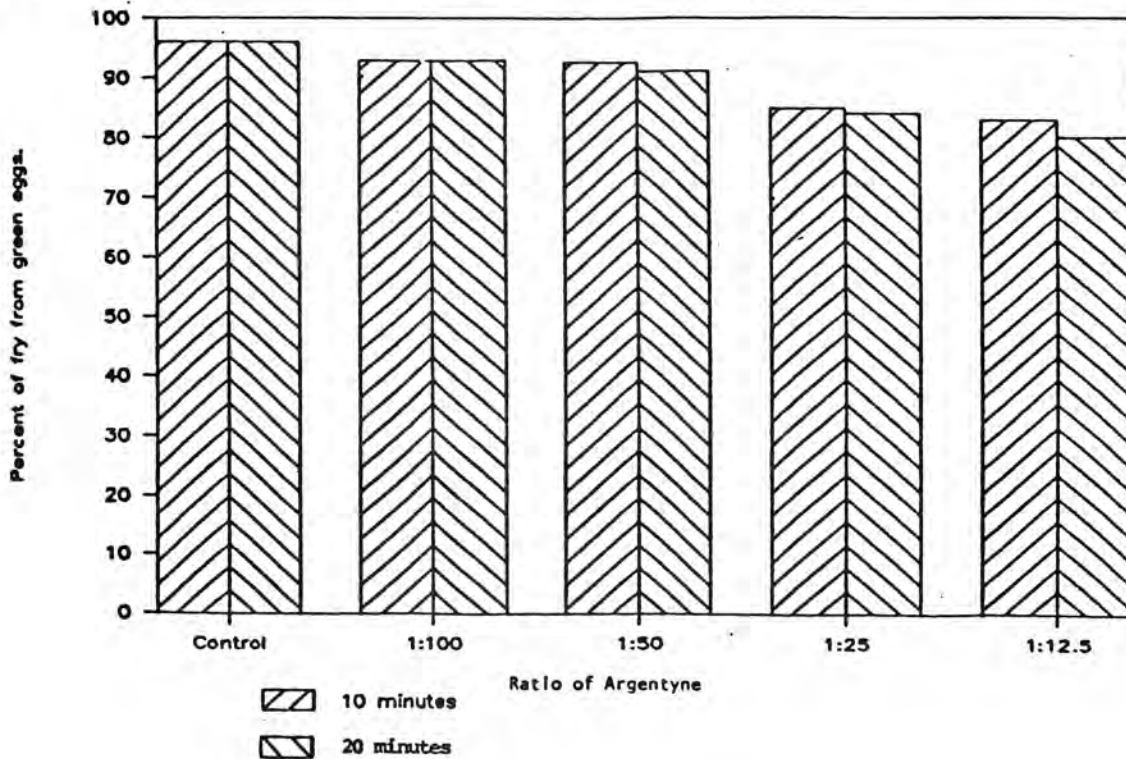
| Set | Argentyne ratio | pH | Treatment time | | Mean | S.D. |
|-----|-----------------|-----|-----------------|---------|------|------|
| | | | 10 min. | 20 min. | | |
| | | | (Percent Fry) | | | |
| 1 | Control | 7.8 | 96 | 96 | 96 | 1.54 |
| 2 | 1:100 | 6.6 | 93 | 93 | 93 | 1.24 |
| 3 | 1: 50 | 7.3 | 92 | 91 | 91.8 | 2.74 |
| 4 | 1: 25 | 7.2 | 85 | 84 | 84.5 | 1.88 |
| 5 | 1: 12.5 | 7.2 | 83 | 80 | 81.5 | 4.42 |

The compressed table of p values is:

| SET | 1 | 2 | 3 | 4 | 5 |
|-----|------|------|------|------|------|
| 1 | X | H.S. | H.S. | H.S. | H.S. |
| 2 | H.S. | X | N.S. | H.S. | H.S. |
| 3 | H.S. | N.S. | X | H.S. | H.S. |
| 4 | H.S. | H.S. | H.S. | X | V.S. |

Where: N.S. = non significant, (= or >0.05), S. = significant (= or >0.02), V.S. = very significant (= or >0.01), H.S. = highly significant (= or >0.001)

Figure 1. Results of egg hardening in iodophore solution.



T-tests showed significant differences between the control group (Set 1) and the treatment groups (Sets 2,3,4, and 5) and between treatment groups, except between the 1:100 and 1:50 Argentyne treatment. However, no significant difference was found between the 10 and 20 minutes treatments.

Increased vitamin C in the egg hardening process seemed to be beneficial up to the

amount of about 0.1 g/L. Because vitamin C is acidic, the effect on pH was buffered with NaHCO₃ to keep the pH readings at 7.4. The addition of vitamin C to the 1:100 Argentyne solution for egg hardening improved fry survival. For the control, 1:100 Argentyne solution without vitamin C was used. No extender or rinse was used in fertilization (Table 2 and Figure 2).

TABLE 2.

Fry survival associated with several concentrations of vitamin C added to 1:100 Argentyne solution used for egg hardening, from 12 observations per set.

| Set | Vitamin C in mg/L | Treatment time | | Mean | S.D. |
|-----|----------------------|----------------------------|---------|-------|------|
| | | 10 min. (Percent fry) | 20 min. | | |
| 1 | Control | 49 | 48 | 48.5 | 2.32 |
| 2 | 20 | 53 | 50 | 51.5 | 3.85 |
| 3 | 40 | 54 | 53 | 53.5 | 2.88 |
| 4 | 80 | 59 | 57 | 58.17 | 2.72 |
| 5 | 160 | 58 | 48 | 53.0 | 8.52 |

The compressed table of p values is:

| SET | 1 | 2 | 3 | 4 | 5 |
|-----|------|------|------|------|------|
| 1 | X | H.S. | H.S. | H.S. | H.S. |
| 2 | H.S. | X | N.S. | H.S. | H.S. |
| 3 | H.S. | N.S. | X | H.S. | H.S. |
| 4 | H.S. | H.S. | H.S. | X | V.S. |

Where: N.S. = non significant, (= or >0.05), S. = significant (= or >0.02), V.S. = very significant (= or >0.01), H.S. = highly significant (= or >0.001)

Significant differences were shown with T-tests between the control (Set 1) and each of the vitamin C treatments, but no significant difference was found between the 20 mg/L and 40 mg/L vitamin C treatments (sample cases of 12 per set).

In additional experiments, eggs were treated the same as above, with the exception that sperm was not introduced to them. Instead the semen still active in the first egg washing solution fertilized 6 % and 1 % of the eggs respectively, in the

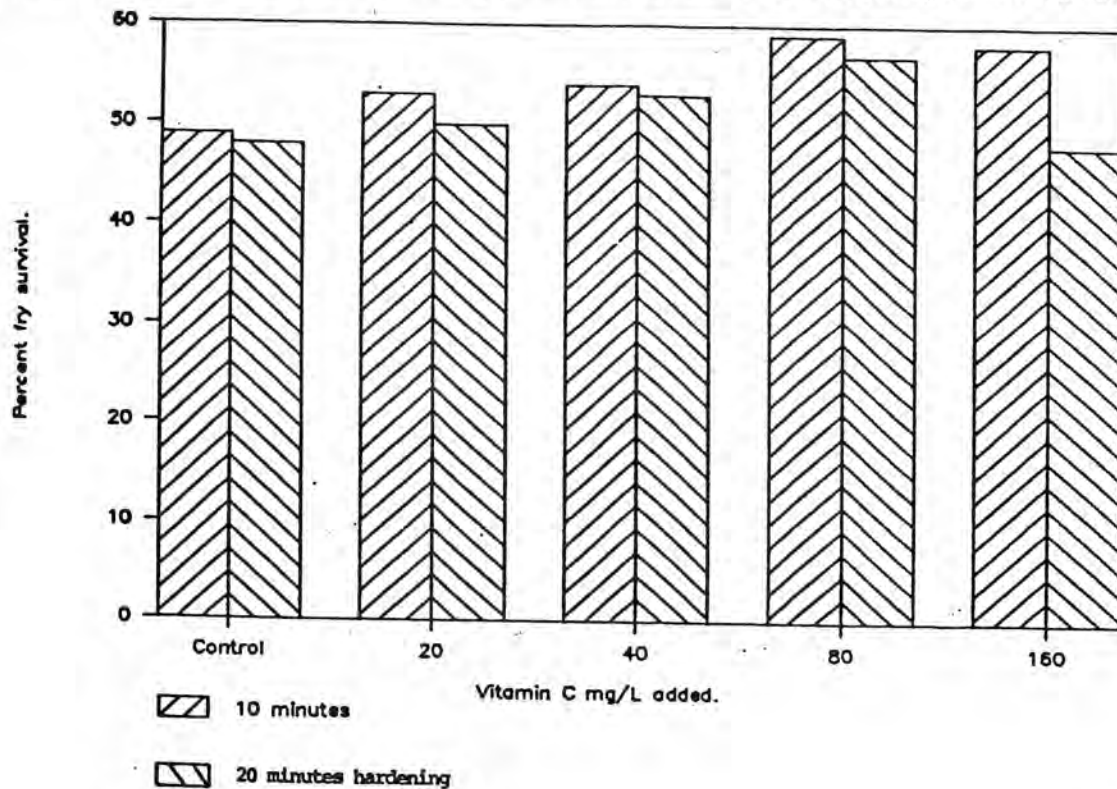
first and second washing.

DISCUSSION

Yoshimizu et al. (1989) doubted the possibility of vertical transmittance of IHNV, as did other authors regarding IPNV (Wechsler et al. 1987).

None-the-less, IPNV is a much tougher virus than either IHNV or viral hemorrhagic septicemia (VHS), which

Figure 2. Fry survival after egg hardening in 1:100 Argentyne solution.



unlike the former are sensitive to ether heat (Amend 1974). Furthermore, IPNV can be transferred with acetone-treated pituitary injections (Ahne 1985).

Presently we can only speculate on the apparently contradictory assessments of these viruses. However, in our hatcheries a group of older fish receiving elevated amounts of vitamin C remained unaffected when probably exposed to IPNV infection. Although we have no direct proof that the vitamin C gave protection against IPNV, there are observations that it can give protection against IHNV (Halver 1989).

Earlier communication with Dr. Halver (1988) led to our experiments with elevated amounts of vitamin C in the egg hardening process. Vitamin C improved fry survival when applied at concentrations up to 1.0g/L (Table 2). Using stable vitamin C would certainly improve the results. The iodophore treatment caused a 2% - 3% decrease in

fry survival (Table 1), but considering the importance of disease prevention, it seems to be a small price to pay.

Certainly there is conflicting information about the survivability of IPNV or other infections and the effectiveness of disinfecting the eggs (Bullock et al. 1976, Elliott et al. 1978). However, Yoshimizu et al. (1988) reported a decrease in *O. masou* virus (OMV) in Japan since iodophore treatment of eyed eggs was introduced. Considering the risk and the seriousness of these infective diseases, no procedure should be ignored that can offer hope in preventing their spread.

The revised procedure for fertilization and egg treatment, which we have developed on the basis of indications from the present study, can be summarized as follows:

1. Check for ripe fish at least once, but not more than twice, a week.

2. Do not keep ripe fish more than three days in cages. If possible, spawn them the day after sorting.
3. While spawning, collect the eggs in a strainer, separating them from the ovarian fluid.
4. Rinse the eggs in a solution of 3% NaCl + 0.05% EDTA, moving the strainer up and down a few times in the solution. After the eggs are rinsed, they should be drained for about one minute. The rinsing solution can be made using holding pond water, preferably from near the inlet, in which the spawning fish had been kept. About 10 L of solution are necessary to rinse 300,000 eggs.
5. Use a dry pan for spawning. Squirt semen from one male into the pan, then introduce the eggs. The eggs from three to five females can be fertilized using two to five males. Mix the eggs and semen immediately after the semen from each male is introduced.
6. Add the oxygen-bubbled extender solution: 6 g/L NaCl, 0.2 g/L CaCl₂, and 0.4 g/L B-glucose. Mix solution with eggs for one minute. Let these stand for about five minutes, but not more than 15 minutes. Distilled or de-ionized water should be used to make the extender solution which can be stored in a cool place for two to three weeks. Before use, the extender solution should be oxygenated by bubbling air (preferably oxygen) through it. The solution temperature should be adjusted to approximately that of the water in which the spawning fish are kept. Use about 50 ml of solution for 3,000 rainbow trout eggs.
7. Thoroughly but gently wash the eggs clean of extender solution and semen by using 30 cm diameter sieves in pond water. We filled two containers with pond water and gave the eggs a first and a second washing. For the first washing, we used an 11.25 mg/L ZnSO₄, 1.0 g/L NaCl, 0.5 g/L

NaHCO₃, and 0.5 g/L vitamin C stock solution in distilled water. We combined one part of this solution to four parts of pond water and added, immediately before use, 1:100 Argentyne. Adjust the pH to 8.0 adding NaHCO₃.

For the stock solution for the second washing, we dissolved 11.25 mg/L ZnSO₄, 1.0 g/L CaCl₂, 0.5 g/L vitamin C, and 0.5 g/L NaHCO₃ in distilled or de-ionized water. Again, we used the solution at a ratio of one part solution to four parts of pond water. The eggs should be washed in a sieve for at least five minutes in each solution. A rope handle can facilitate working with the sieves. Change the water after washing 100,000 eggs.

8. Harden the eggs in a solution of 0.45 mg/L ZnSO₄, 0.04 g/L CaCl₂, 0.02 g/L NaCl, and 0.1 g/L vitamin C for one to two hours. The egg hardening water should be saturated with oxygen and ammonia-free.
9. Treat the eggs with 1:300 iodophore (Argentyne or Ovadine) for 10 minutes before they are placed in the incubators.

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For their support and suggestions, I would like to thank Dr. Halver, University of Washington; the hatchery managers and assistant managers- Red (John) Barnhardt, Winfried Schenk, Rod Burns, and Dave DePape; my supervisors and director M. Drouin, T.W. McFadden, and T. Mill; the hatchery staff- J. Bilas and D. Publack; and last but not least to Dave Ealy for his help in editing.

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**STEELHEAD EGG HARDENING IN VITAMIN
C-3 IMPROVES HATCHABILITY**

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Abstract

Replicate lots of 500 steelhead trout eggs spawned from three females and fertilized with combined milt from two males were water hardened during a one hour treatment in 0, 50, 500, or 1,000 ppm of Stay-C (vitamin C₃ plus small amounts of mono- and diphosphate derivatives). Samples of developing eggs were removed at days 1, 20, and 40, and at hatch and were analyzed for content of L-ascorbic acid (C₁), L-ascorbyl-2-sulfate (C₂), and L-ascorbyl-2-triphosphate (C₃). Daily mortality was recorded for each lot held in screen-bottom sample rings in one Heath incubator tray supplied with 12°C water in the School of Fisheries, University of

Washington hatchery. Development egg mortality was low except in highest C₃ treatment hardening. Hatch was directly related to C₃ water hardening treatment as follows: 0 ppm: 84%; 50 ppm: 96%; 500 ppm: 90%; 1,000 ppm: 43%. Therefore, C₃ at 50-500 ppm in water for water hardening improved hatch and survival of steelhead trout eggs.

Direct HPLC assay for vitamin C using 5% TCA extraction technique and both EC and UV detectors failed to detect enhanced levels of C₁, C₂, or C₃ in the egg or yolk-sac fry tissue.

This study was supported by Rangen, Inc., Buhl, ID.

DOES GRADING FISH REDUCE OVERALL SIZE VARIATION?

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ABSTRACT

To determine if grading would reduce overall size variation in fish population a grading study was initiated. Summer steelhead were hand graded producing two raceways of small fish and two raceways of large fish. Two ungraded raceways served as control. Fish were fed a varying amount as determined by computerized growth program to produce the same size fish at the completion of the production cycle. Two hundred fish were measured from each raceway at the 0, 90, and 180 days later. Grading was ineffective in reducing the overall size variation in the summer steelhead population.

INTRODUCTION

Fish are sometimes graded in a hatchery to obtain more uniform size fish for processing, distribution, or to increase the efficiency of the hatchery. Increasing the efficiency is based on the assumption that smaller fish will increase their growth rate when segregated from a mixed size population. Past studies (Pyle et al., 1961; Pyle, E. A., 1964; Pyle E.A., 1966) have shown the above assumption to be

incorrect. However, the above studies did not address whether a overall reduction in fish size variation was seen in fish population which were graded and reared separately. The objective of this study was to test whether grading fish would reduce the overall size variation in a fish population as compared to an ungraded population.

Methods

The experiment was conducted at Chelan Hatchery, Washington Department of Wildlife located at Chelan Falls, Washington. The fish used were 1988 brood year Wells stock summer steelhead. The fish were reared in 10 ft by 100 ft by 3 ft concrete raceways supplied with well and spring water which averaged 54 to 56 F during the experiment. The fish were fed closed formula commercial feeds using demand feeders.

In September, the fish were hand graded with a bar grader into two groups, small or large and distributed into the raceways according to size. Control fish were not graded. The fish were distributed in the raceways as per the following table:

Table 1. Distribution of fish in raceways.

| Test Group | Raceway No. | Fish Size No./lb | Fish No. |
|------------|-------------|---------------------|----------|
| Small 1 | 6 | 76 | 18,200 |
| Small 2 | 7 | 76 | 18,300 |
| Large 1 | 2 | 34 | 14,400 |
| Large 2 | 5 | 34 | 15,000 |
| Ungraded 1 | 4 | 54 | 24,400 |
| Ungraded 2 | 8 | 54 | 24,400 |

Two hundred fish from each were randomly selected, anesthetized in MS-222, and lengths measured on October 3, 1988, January 3, 1989, and April 3, 1989.

Fish were fed using a Lotus 1.2.3 worksheet which projected growth for a 3 month period. Fish were fed to produce the same average size fish in all groups at the completion of the production cycle.

RESULTS AND DISCUSSION

At the beginning of the study, grading the fish did results in two raceways of fish with average fish length of 88 mm as compared to 111 and 113 mm for the large fish (Table 1). At the completion of the study the fish in all raceways were comparable in average fish length (Table 2). Graphic representation of this data is shown in Figure 1.

Table 2. Average fish length, graded and ungraded fish.

| Test Group | Average Fish Length (mm.) | | |
|------------|---------------------------|--------|--------|
| | Oct. 3 | Jan. 3 | Apr. 3 |
| Small 1 | 88 | 152 | 185 |
| Small 2 | 88 | 149 | 190 |
| Large 1 | 113 | 160 | 198 |
| Large 2 | 111 | 159 | 197 |
| Ungraded 1 | 102 | 160 | 195 |
| Ungraded 2 | 101 | 156 | 198 |

Figure 1. Average fish length, graded and ungraded fish.



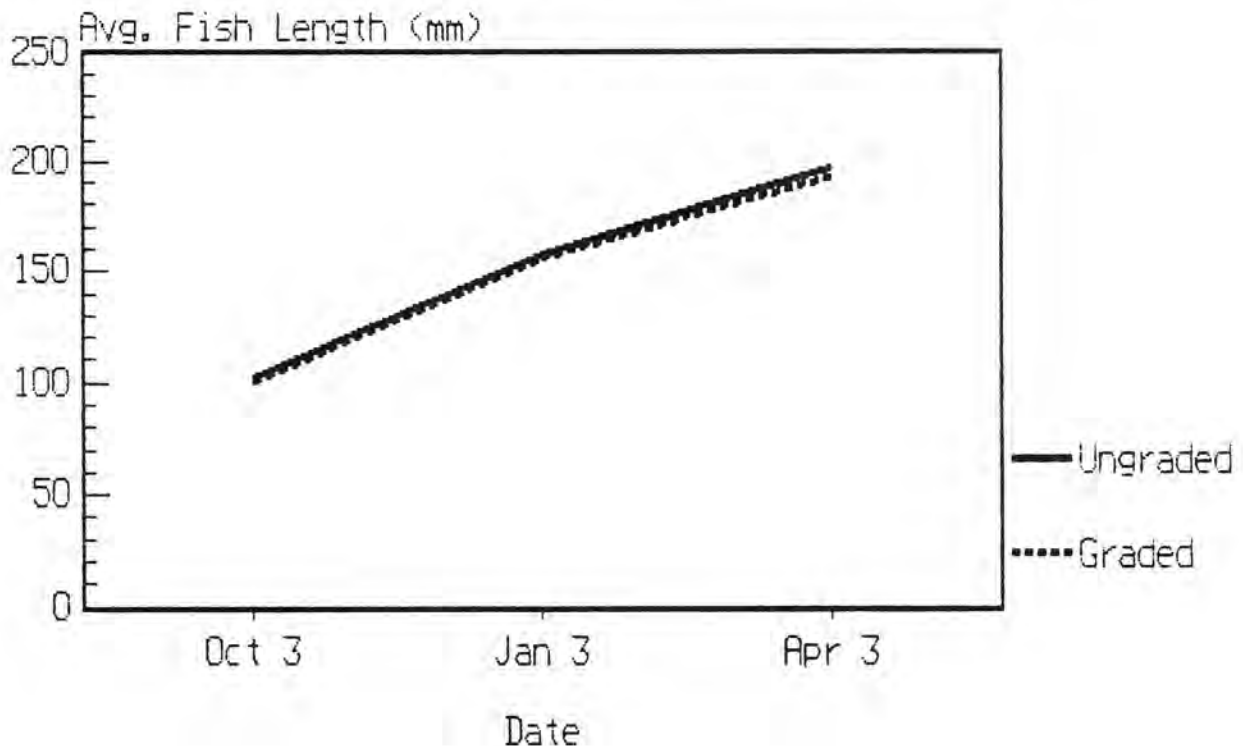
However when the data was combined for all graded fish versus ungraded fish the average size are very comparable at the

beginning, middle, and end of the study (Table 3). Graphic representation of this data is shown in Figure 1.

Table 3. Average fish length, combined graded versus ungraded fish.

| Test Group | Average Fish Length (mm.) | | |
|------------|---------------------------|--------|--------|
| | Oct 3. | Jan. 3 | Apr. 3 |
| Graded | 100 | 155 | 193 |
| Ungraded | 102 | 158 | 197 |

Figure 2. Average fish length, combined graded versus ungraded fish.



The standard deviation is a statistic that measures the variation of a set of numbers around their mean. However, statistically it is not possible to compare the standard deviations of two groups if they have different means. The coefficient of

variation allows for the comparison of the variability of two groups which have different means.

The coefficient of variation for the combined data of the graded fish was lower

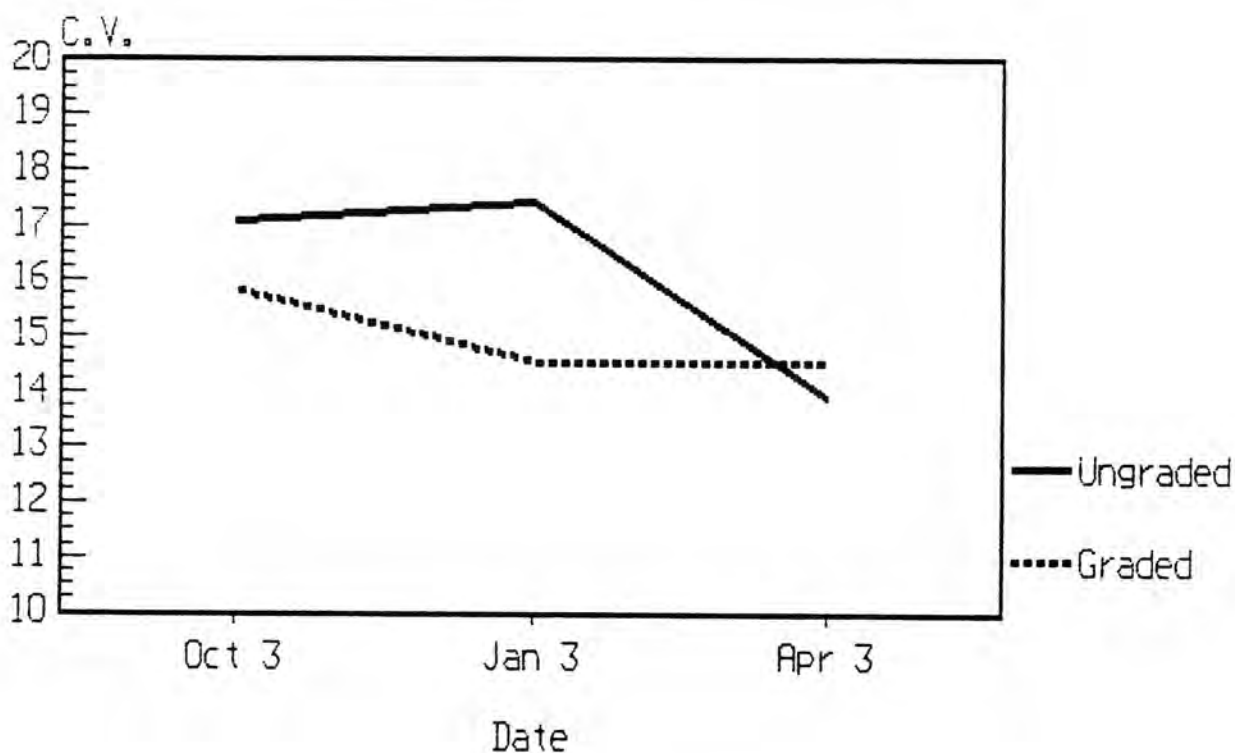
then the ungraded fish at the begin of the study (Table 4). At the completion of the study, 180 days later, the coefficient of variation for the graded fish was slightly higher then the ungraded fish (Table 4).

Graphic representation of the coefficient of variation for the combined data for graded and ungraded fish are seen in Figure 3.

Table 4. Coefficient of variation, combined graded versus ungraded fish.

| Test Group | Coefficient of Variation (%) | | |
|------------|------------------------------|--------|--------|
| | Oct 3. | Jan. 3 | Apr. 3 |
| Graded | 15.8 | 14.5 | 14.5 |
| Ungraded | 17.1 | 17.4 | 13.9 |

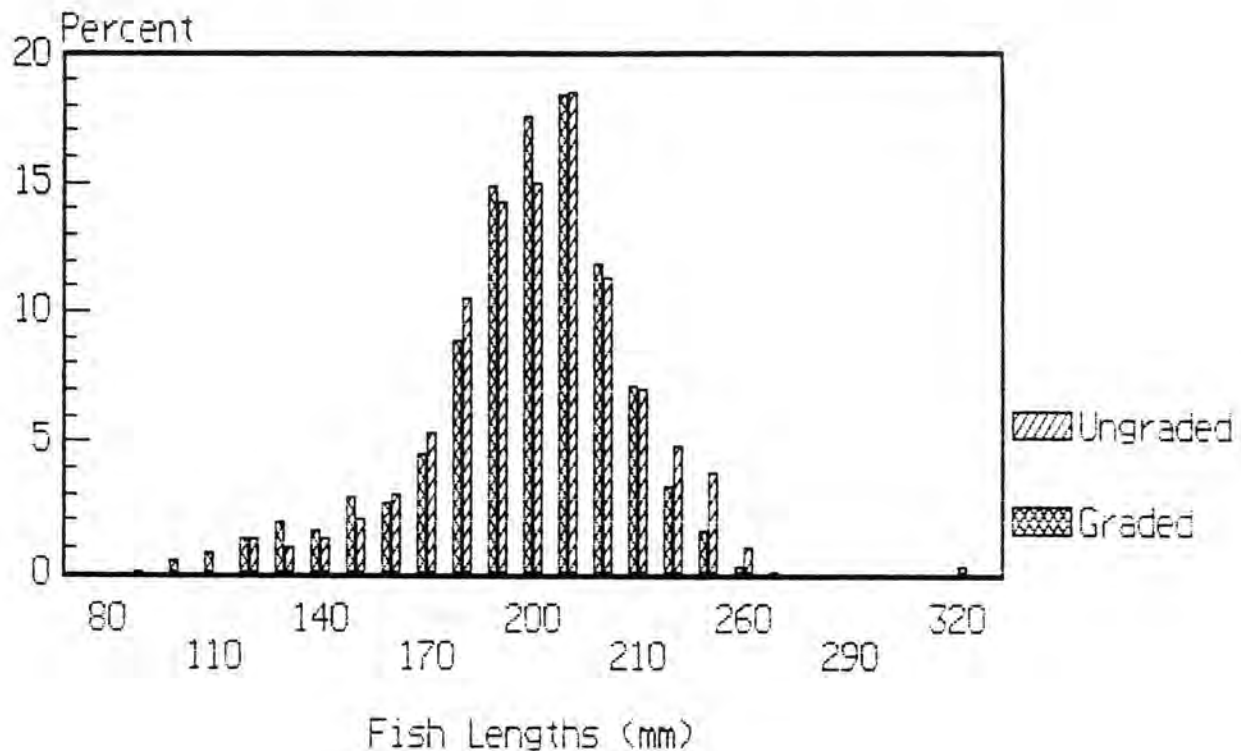
Figure 3. Coefficient of variation, combined graded versus ingraded fish.



Plotting a length frequency histogram also showed that at the completion of the study

both graded and ungraded were comparable in size distribution (Figure 4).

Figure 4. Fish length frequency histogram, graded versus ungraded fish.



CONCLUSIONS

Grading fish was ineffective in reducing the overall size variation in a fish population. If an adequate diet in terms of quality and quantity are fed to a group of fish the amount of size variation in the population is primarily a function of the genetic variation.

Thus, the value of grading to reduce overall size variation is questionable. However, grading to remove fish for processing as in a commercial food fish hatchery is not questioned.

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DEMAND FEEDERS: DOES THE NUMBER OF FISH PER FEEDER AFFECT GROWTH PERFORMANCE?

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INTRODUCTION

Demand feeders have been used at only a few WDF hatcheries. Recent studies at the Humptulips salmon hatchery have focused on using demand feeders to regulate growth of coho salmon (*Oncorhynchus kisutch*) in both raceways and 0.5 acre rearing ponds containing as many as 900,000 fish. A recent study (Fuss et. al., 1988) at the hatchery compared food conversion and feed efficiency rates, condition factors, and size variation of coho salmon reared in raceways and fed a weeks ration in either 3 or 7 days. In these studies, only 2 demand feeders were used to feed 40,000 (20,000 per feeder) coho.

Prior to the initiation of these studies, several commercial trout growers and public agency hatcheries were contacted to determine if there was a recommended number of fish/feeder. There was no agreed upon number, and in-use numbers ranged between several hundred up to about 10,000 fish/feeder. The importance of number of fish per feeder is not known. Conceivably, too many fish per feeder could result in increased competition for food at low ration levels. This could result in increased size variation and a decrease in growth performance. Also, the loading rate of the rearing vessel should be taken into consideration when determining how many feeders to use as too few feeders could cause unhealthy densities of fish around the feeders.

The following study was conducted at the Humptulips Salmon Hatchery to determine if a relationship existed between number of fish per feeder and food conversion rates, feed efficiency, percent weight gain, and size variation.

METHODS

Approximately 40,000 fish were stocked in each of four 20 X 80 foot concrete

raceways. Size at stocking varied among the raceways and ranged between 111 and 114 fpp. Three treatments (Figure 1) denoted as # of feeders per raceway (#PR) were established and compared to a control. The treatments were 6PR (6,667 fish/feeder), 4PR (10,000 fish/feeder), and 2PR (20,000 fish per feeder). The control was a single raceway containing 40,000 fish which were hand fed. Starting on July 1, 1989, fish were fed a fixed ration of Clarks New Age Diet #3 pellet. Rations were adjusted monthly according to a predetermined growth schedule. Fish were sampled semimonthly during the fall. The mean length, weight, coefficient of variation (CV), and condition factor were computed from a sample of 100 fish. Food conversions, feed efficiencies, and growth rates (mm/day) were calculated between sampling periods and for the overall duration of the study. Two months into the study, low food conversions indicated that a population shortage existed in the control group raceway. As a result, this raceway received a greater feed ration than the treatment groups during this period. Consequently, the growth rate and percent weight gain were higher for the control group.

RESULTS

There were no apparent differences in food conversions (Figure 2) among the groups. The control group had a slightly lower food conversion than the treatment groups. The 6PR and 2PR had the highest food conversions.

The control group also had the highest feed efficiency (Figure 3). The 6PR and 2PR had the lowest feed efficiencies.

The percent gain in weight was similar among the treatment groups (Figure 4). The control group had the highest percent gain because of the previously mentioned problem.

Size variation as measured by the coefficient of variation (CV) did not vary greatly (Figure 4) among the groups nor did it change greatly between the beginning and end points of the study. Size variation in all groups was well within acceptable limits of WDF criteria.

Growth rates among the treatments were similar but lower than the control. This is due to the control being fed at a higher ration level than the treatments.

DISCUSSION

This study sought to determine whether food conversion, feed efficiency, percent weight gain, and size variation was affected by the number of fish per demand feeder.

At the stocking levels used in this study, there were no discernable differences among groups in the parameters evaluated. Although the treatments did not perform quite as well as the control group, these differences were so slight we judge them to be inconsequential.

We have fed as many as 112,000 fish per feeder in large rearing ponds (WDF, unpublished) and 20,000 per feeder in

raceways (Fuss and Seidel, 1988) with acceptable results. The levels used in this study were matched or exceeded those used at some public agency production hatcheries and commercial trout operations.

In this study, the number of fish per feeder ranged between 6,700-20,000 and ration levels were above maintenance. Whether similar results would occur at limited ration levels was not determined.

ACKNOWLEDGEMENTS

We wish to extend our sincere thanks to the crew at the Humptulips Salmon Hatchery for their cooperative spirit and attention to detail. We would also like to thank Kent Dimmitt, Andy Appleby, and Mark Kimbel for their assistance in collecting data.

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Fuss, H.J., P.R. Seidel, K.R. Dimmitt, and A.E. Appleby. 1988. Feeding strategies for coho salmon using demand feeders. Proc. 39th Annual NW Fish Culture Conference, Richmond, B.C.

NUMBER OF FISH PER DEMAND FEEDER # FEEDERS PER RACEWAY 2-6

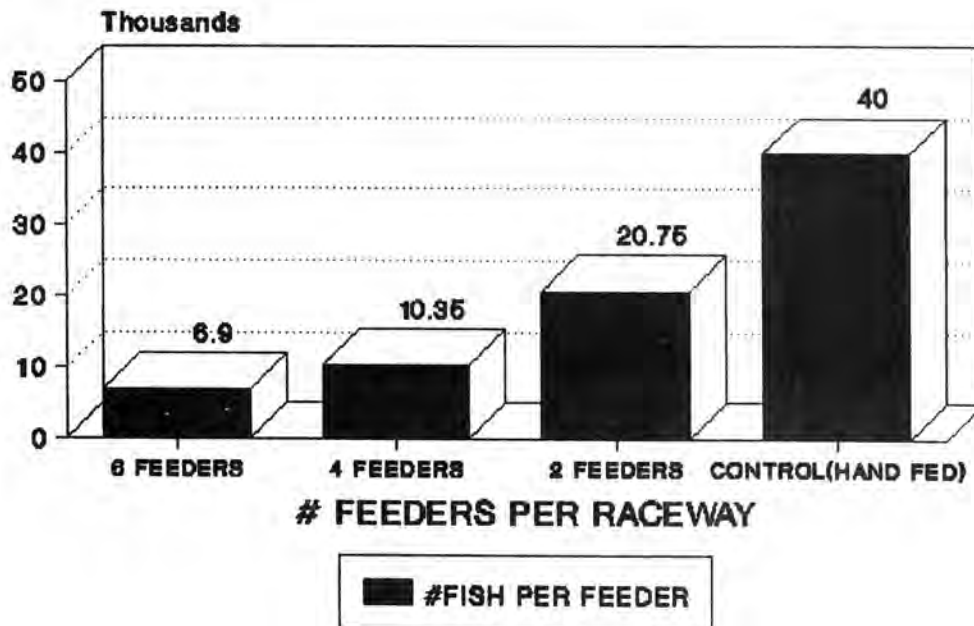


Figure 1.

FOOD CONVERSION

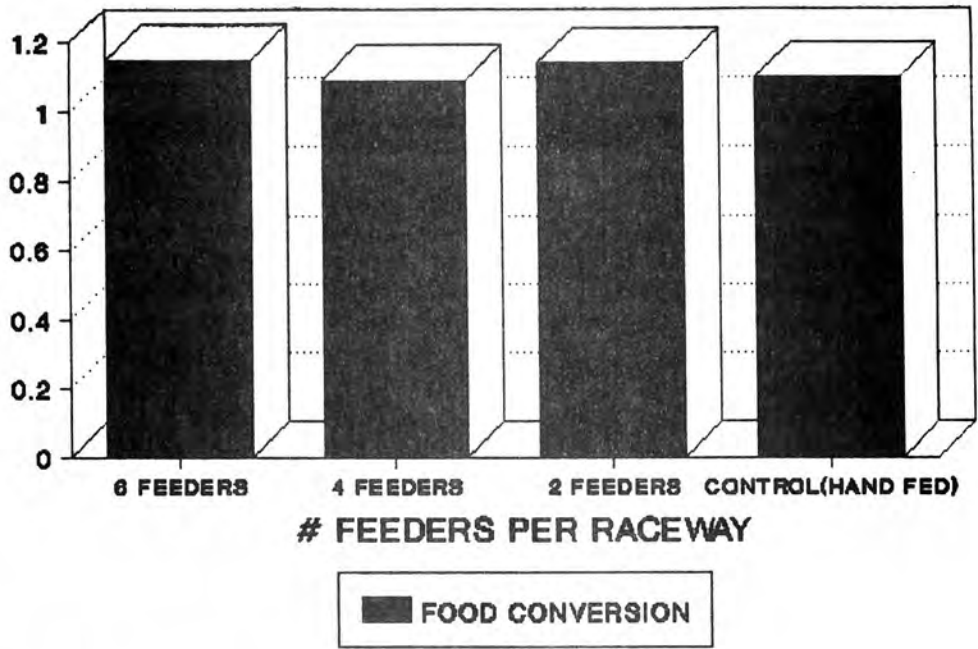


Figure 2.

FEED EFFICIENCY

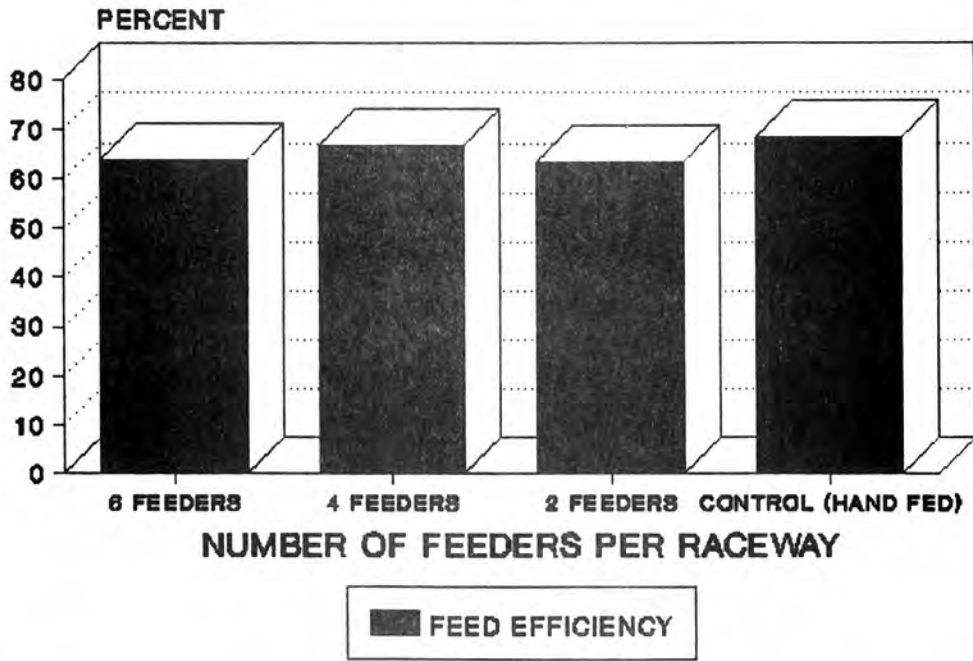


Figure 3.

PERCENT WEIGHT GAIN

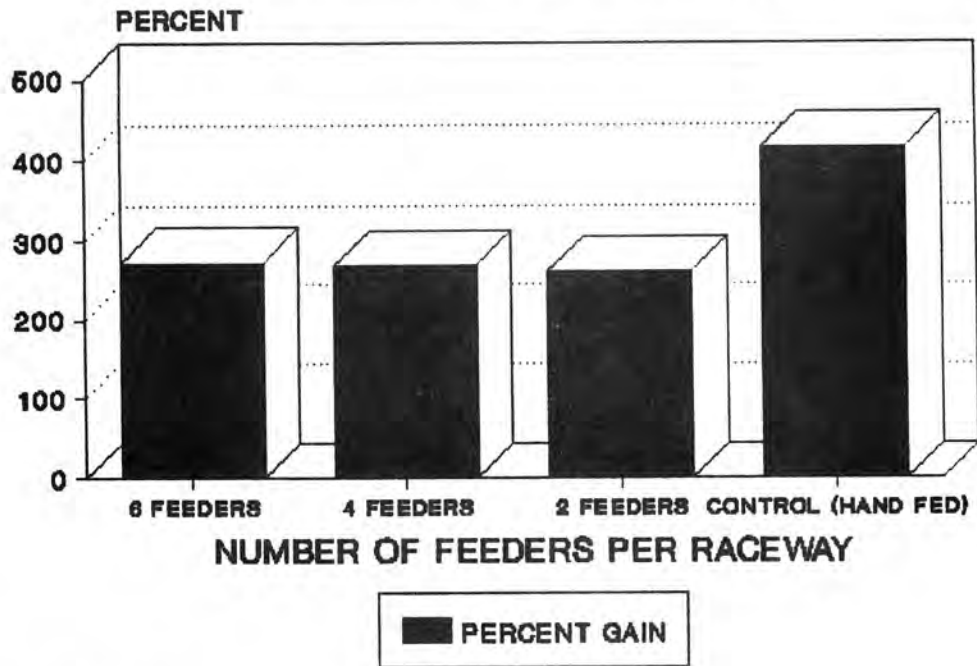


Figure 4.

SIZE VARIATION (CV) BEGINNING AND END

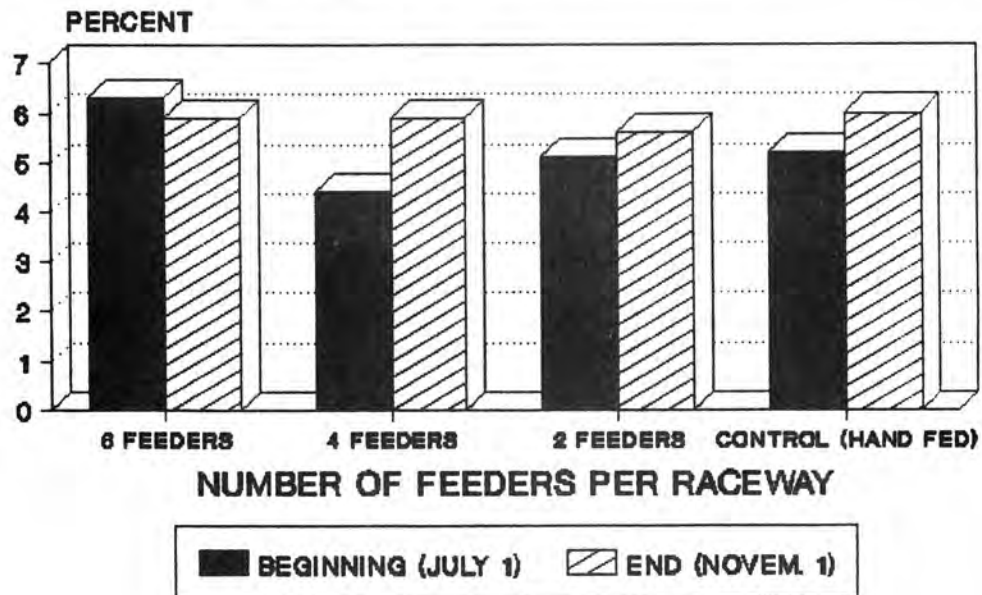


Figure 5.

**RESULTS AND ECONOMICS OF THE 1989 HARDY SEAFARMS-BIOMED FIELD TRIALS:
RE-VACCINATION OF CHINOOK SALMON IN SEAWATER TO PREVENT VIBRIOSIS**

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Most chinook salmon farms vaccinate their fish in the fresh water hatcheries by direct immersion in diluted vaccine to prevent vibriosis disease after they are transferred to the marine pens. This usually occurs in late March or early April in 8 to 11°C water. The fish are transferred to the marine net-pens three to four weeks later in order to allow a minimum of two weeks to develop immunity before taking a chance on exposure to vibrios.

Unfortunately, the duration of the immune response to single immersion vaccination does not appear to be as good in chinook salmon as it is in coho or Atlantic salmon or rainbow trout. An example from earlier work done in Puget Sound by scientists from TAVOLEK, Inc. demonstrates this perfectly. In this case, they used "Relative Percent Survival" or RPS, which compares the test group mortality against a control group that is usually never medicated or vaccinated. Immersion vaccinated coho had over a 90% RPS that lasted for over 300 days, compared to about an 85% RPS for chinook that peaked after 50 days. The low point of RPS was after about 125 days, and came up slightly after that. All of these fish were held in fresh water and sub-sampled periodically for challenge tests. In the "real world" (that is, the marine net-pens), the chinook are getting a natural vibrio challenge every day from May through winter. These challenges usually begin with the common, "warm-water" vibrio, *Vibrio anguillarum*, in

the spring, followed by the appearance of the "cold-water" vibrio, *Vibrio ordalii*, in late summer. On chinook salmon farms, we find that the peak of protection to both of these vibrios (or "serotypes") is somewhere between 80 and 120 days post immersion vaccination, and then begins to wane. Zero-age chinook vaccinated in fresh water in mid-April begin losing immunity in late June, just when protection is needed the most. This varies with size of the fish at vaccination, marine water temperatures, stress and presence of vibrios. Larger fish are more immunocompetent, and BIOMED prefers to postpone vaccinating until the fish average 5 grams whenever possible. We also prefer to immersion vaccinate when the water temperatures are at least 8°C and the fish are not being stressed by other diseases or smolting. Prolonging immunity as long as we can may become more important, as BIOMED diagnosticians are finding an increased incidence of both serotypes in mortalities examined from both the northern Puget Sound and Southern B.C. farms.

Each summer, Hardy SeaFarms has had to put vaccinated fish on antibiotics. We knew that we didn't want to give the fish that "primary", or first vaccination when they were in the sea-pens, as first vaccinations in the presence of the live organism can actually create the problem rather than cure it. But, the Norwegian veterinarians had reported excellent success re-vaccinating Atlantic salmon in sea-pens,

and this was the direction that we decided to go with a field trial. We decided to try a comparison of immersion re-vaccination in Ocean Farms fresh water hatchery at Duncan and in the marine net-pens at the Hardy SeaFarms Tank Farm site on Hardy Island, plus re-vaccination at the Tank Farm by using the Norwegian bath techniques. The main difference between immersion and bathing is that immersion is usually a 20 to 30 second dip in an aerated dilution of the vaccine, and bathing is usually 30 to 60 minutes in an aerated vaccine bath diluted 50 to 100 times more than the immersion. The latter method is supposed to be less stressful on the fish, but requires more vaccine to be effective. The main thing was that the fish had to have that first "primary" vaccination at the fresh water hatchery to be safe. That was the key element in all other decisions.

We decided that if we could "re-vaccinate" these fish in the salt water pens in May, before the vibrio season begins, survival would improve and the need for using medicated feeds would be reduced.

The permutations were such that we collected data on a total of 12 pens: 6 production units and 6 research sized pens. The production units had populations ranging from 15 to 29,000 fish each, and the research units started with about 6,000 fish each. We could not risk putting any

totally "naive" (that is, fish that were never vaccinated) fish in the water without using oral medication, but we did want to make a direct economic comparison with the cost of medicating fish that had had just a single, primary vaccination in fresh water. Our "control" comparison was a production pen (Tank Farm Pen #11) that was given a primary immersion vaccination of a commercial B.C. vaccine which was a blend of both vibrio serotypes on March 29, 1989 at an average size of 4 grams, and an immersion re-vaccination at the hatchery with the commercial B.C. *Vibrio ordalii* vaccine only on April 13, 1989 when the fish averaged 6.2 grams. These fish were never medicated at the Tank Farm net-pen site. 28,880 fish were helicoptered to the pen on April 18, 1989.

Our "medicated" comparison was production Pen #13. These fish were given a single, primary immersion vaccination at the Duncan hatchery with the same commercial B.C. vaccine mixture of the two vibrio strains on 29 March, 1989 when they averaged 6 grams. 14,812 fish were helicoptered to the pen on 14 April, 1989 when they averaged 8.7 grams. Fish in Pen #13 were medicated as needed during vibriosis epizootics.

At the end of September, this is what the actual mortalities in these two pens looked like:

| PEN NO. | DATES AND CUMULATIVE PERCENT(%) MORTALITY | | | | |
|--|---|---------|---------|---------|---------|
| | 5/31/89 | 6/30/89 | 7/31/89 | 8/31/89 | 9/30/89 |
| <u>11</u> controls [primary and re-vaccination in fresh water with B.C. vaccines] | 0.56 | 12.63 | 17.76 | 18.34 | 19.59 |
| <u>13</u> medicated controls [primary vaccination only; medicated as needed] | 0.63 | 4.10 | 8.20 | 9.60 | 11.10 |

Note that the first big surge in mortalities occurs in June, and that the mortalities began to level off dramatically after the end of July. The medication helped, but a more likely scenario is that the immunocompetant fish survived in the non-medicated group, and the medication helped some of the "border-line" fish survive until they developed a higher degree

of immunocompetence.

So, in a worse case situation, it appears that immersion vaccination and re-vaccination in fresh water with no medication in salt water will account for about a 20% mortality through September, and about 11% with medication.

We also conducted tests with primary immersion and re-vaccination in fresh water using BIOVAX ^{1/}.

On April 27, we "BIOVAXed" 5 gram fish with a primary immersion vaccination for Tank Farm Pen #10 (20,355), and on May 11, we "BIOVAXed" 7 gram fish with a

primary immersion vaccination for Tank Farm Pen #2 (20,801). On the same dates, we "BIOVAXed" 5 gram fish with a primary immersion vaccination and re-vaccination for Tank Farm Pen #1. All of these fish were on the same medication program as Pen #13, and this is what the mortalities looked like:

| PEN NO. | DATES AND CUMULATIVE PERCENT(%) MORTALITY | | | | |
|--|---|---------|---------|---------|---------|
| | 5/31/89 | 6/30/89 | 7/31/89 | 8/31/89 | 9/30/89 |
| <u>10</u> FW BIOVAX primary (4/27/89) | 0.31 | 2.13 | 7.88 | 9.01 | 10.20 |
| <u>2</u> FW BIOVAX primary (5/11/89) | 0.25 | 1.55 | 10.52 | 11.20 | 11.90 |
| <u>1</u> FW BIOVAX primary (4/27/89) and fresh water re-vaccinate (5/11/89) | 0.37 | 1.40 | 5.92 | 6.77 | 7.54 |

and, if we look at Relative Percent Survival (RPS) where compared against CONTROL PEN=#11 we find:

| PEN NO. | RELATIVE PERCENT SURVIVAL ^{2/} | | | | |
|----------------------|---|---------|---------|---------|---------|
| | 5/31/89 | 6/30/89 | 7/31/89 | 8/31/89 | 9/30/89 |
| PROD. <u>PEN #10</u> | 44.64 | 83.14 | 55.63 | 50.87 | 47.93 |
| PROD. <u>PEN # 2</u> | 55.36 | 87.73 | 40.77 | 38.93 | 39.25 |
| PROD. <u>PEN # 1</u> | 33.93 | 88.92 | 66.67 | 63.09 | 61.51 |
| PROD. <u>PEN #13</u> | -12.50 | 67.54 | 53.83 | 47.66 | 43.34 |

There was a definite improvement by re-vaccinating in fresh water, but this means holding the fish in the fresh water hatcheries longer than economics permit. Could this situation be improved by re-vaccinating after the fish have gone into the net-pens?

We conducted re-vaccination tests with BIOVAX immersions and baths in salt water, both with and without a 5 day post-vaccination oral medication with ROMET 30. Even though the 30 minute "bath" in one of the production pens was very successful, the procedure was complex,

^{1/} BIOVAX is the trade name of BIOMED Inc.'s U.S.D.A. and C.M.A. (Canadian Ministry of Agriculture) licensed bivalent immersion vaccine for both *Vibrio anguillarum* and *Vibrio ordalii* in a single product.

^{2/} where RPS= 1-[%TEST MORTALITY/%CONTROL MORTALITY]*100

labor intensive and the best results were four times more costly per survivor than salt water immersion re-vaccination, and three times more costly than Pen #13 (medicated). Therefore, we will confine this report to the "dip" (30 second immersion) tests.

The BIOVAX immersion re-vaccinations in salt water went very well. These tests were conducted in the research pens. Fish were taken from production PENS 4, 11 and 13 for these tests. Re-vaccinations took place on May 9 and May 18, when we hoped to be before the first vibrio epizootics by several weeks. Dissolved oxygen levels were excellent during the entire season, but we were concerned about surface temperatures, as they were 16 to 18°C! We felt that the stress might be severe, and we set up comparative groups, some with five days of post re-vaccination medication, and some with no medication. No medicated feeds were used in these pens (except as just stated) throughout the season. The differences were insignificant. Several of the pens were hit hard by the *Heterosigma* sp. blooms in September, so for purpose of this report, we'll just discuss two of the non-medicated groups, Pen #C and Pen #F. In all cases, the procedure was as follows:

Large plastic garbage cans were set up with oxygen diffusers right on the perimeter floats of the net-pen to be worked, and for

test data gathering purposes, we set the can on a platform scale. BIOVAX was diluted with straight seawater (which was about 25 ‰ at the time) at a ratio of 1 liter of BIOVAX to 9 liters of seawater until we had at least 30 liters of volume to work with. A knotless nylon bag net was placed in the aerated BIOVAX solution. We crowded the fish into one side of the pen, and "split" the pen by passing a line under it and pulling the bottom above the surface. Fish were dipped from one side into the BIOVAX bag until we had about 10kg, held for 30 seconds and then drained and placed in the "open" side of the pen. The standard licensed usage is 100kg of fish per liter of BIOVAX. After we finished each pen, the lines were dropped and the pens were put back into normal use. Post-vaccination losses over the next few days were amazingly light. Vaccination rates were the same as in fresh water, which are 600kg of fish per hour for a two-person team.

Pen #C fish came from production Pen#13 (see the data above) and were re-vaccinated with BIOVAX in the Tank Farm pen on May 9. Pen #F fish came from production Pen #4, which were given a primary immersion at Duncan hatchery in a mix of the commercial B.C. vaccines of the two vibrio strains on April 5 at 5 grams. These fish were helicoptered to Pen#4 at 7 grams on May 4. The Pen#F fish were re-vaccinated with BIOVAX on May 18. Here are the results:

| PEN NO. | DATES AND CUMULATIVE PERCENT(%) MORTALITY | | | | |
|----------------|---|---------|---------|---------|---------|
| | 5/31/89 | 6/30/89 | 7/31/89 | 8/31/89 | 9/30/89 |
| RES.#C-SW 5/9 | 0.22 | 2.18 | 5.43 | 6.20 | 6.63 |
| RES.#F-SW 5/18 | 0.10 | 1.62 | 5.03 | 5.94 | 7.46 |

and, if we look at Relative Percent Survival compared against CONTROL PEN=#11 we find:

| PEN NO. | RELATIVE PERCENT SURVIVAL ^{2/} | | | | |
|---------------|---|---------|---------|---------|---------|
| | 5/31/89 | 6/30/89 | 7/31/89 | 8/31/89 | 9/30/89 |
| RES.#C | 60.71 | 82.74 | 69.43 | 66.19 | 66.16 |
| RES.#F | 82.14 | 87.17 | 71.68 | 67.61 | 61.92 |
| PROD. PEN #13 | -12.50 | 67.54 | 53.83 | 47.66 | 43.34 |

Comparing these with the medicated production PEN #13 and re-vaccination in the fresh water hatchery PEN #1 shows that re-vaccinating in salt water with BIOVAX is clearly superior to medicating (as shown in the accompanying graph), and will perform as well re-vaccination in fresh water.

Since we have an accurate measure of every activity, including labor, we can make economic comparisons.

Here is an example of a typical cost analysis in our final report for Pen #F:

| | | PEN NO. MAY | JUNE | JULY | AUGUST | OCTOBER |
|------------------------------|---|-------------|--------|--------|--------|---------|
| Starting Number of Fish | F | 6178 | 6078 | 5877 | 5811 | 5717 |
| Monthly Mortality | F | 6 | 94 | 201 | 66 | 94 |
| Cumulative Survivors | F | 6172 | 6078 | 5877 | 5811 | 5717 |
| Avg.Wt.(g) at vaccination | F | 10.1 | | | | |
| Total Wt.-Kgs | F | 62.40 | | | | |
| BIOVAX cost at C\$85.50/L | F | \$53.35 | | | | |
| Man-hours required | F | 0.208 | | | | |
| Labor cost @ C\$7.00/hour | F | \$1.46 | | | | |
| ROMET 30 used - kgs | F | | 0 | 0 | | |
| ROMET 30 cost @ C\$103/kg | F | | \$0.00 | \$0.00 | | |
| Tribrissen used - kgs | F | | | 0 | | |
| Tribrissen cost @ C\$159/kg | F | | | \$0.00 | | |
| Tribrissen feed used - kg | F | | | 0 | 0 | |
| Tribr. feed cost @ C\$522/mt | F | | | \$0.00 | \$0.00 | |
| Monthly cost of medication | F | | \$0.00 | \$0.00 | \$0.00 | |
| Total cost of medication | F | | | | | \$0.00 |
| Cost of drugs+vaccination | F | \$54.81 | | | | |
| Avg. wt. at month end (g) | F | 16.1 | | | 120 | 205 |
| Biomass at month end (kg) | F | 99.4 | | | 697.3 | 1172.0 |
| Cumulat. total cost/kg fish | F | | | | \$0.08 | \$0.05 |
| Month end biomass gain (kg) | F | | | | 598.0 | 1072.6 |
| Cost/kg of biomass gain | F | | | | \$0.09 | \$0.05 |
| Cumulat.cost/1000 survivors | F | | | | \$9.43 | \$9.59 |

* NOTE: the bottom two lines, and the bottom two lines from Pen #13, which is analyzed in the next table. NOTE: also that drug costs include C\$170/mt feed extra for feed preparation.

The cost per kg of biomass gain is about the same, the cost per 1000 survivors is lower for the "BIOVAXed" fish, and no medicated feed was used. A major saving in costs is the reduction in the number of smolts required to achieve a population number by the first fall in the seawater pens, as mortalities could be expected to be approximately 7,000/100,000 smolts for chinook re-vaccinated in the sea-pens, 11,000/100,000 smolts for chinook with

single fresh water vaccinations and medicated as needed, and 20,000/100,000 smolts for chinook with single fresh water vaccinations and no medication during the summer. Chinook smolts cost approximately C\$.40 each. The cost of re-vaccinating in the sea-pens is less than C\$0.01/surviving smolt, compared to C\$0.012/surviving smolt of medicated fish, and approximately C\$1600/100,000 smolts would be saved in capital costs for smolts that survived.

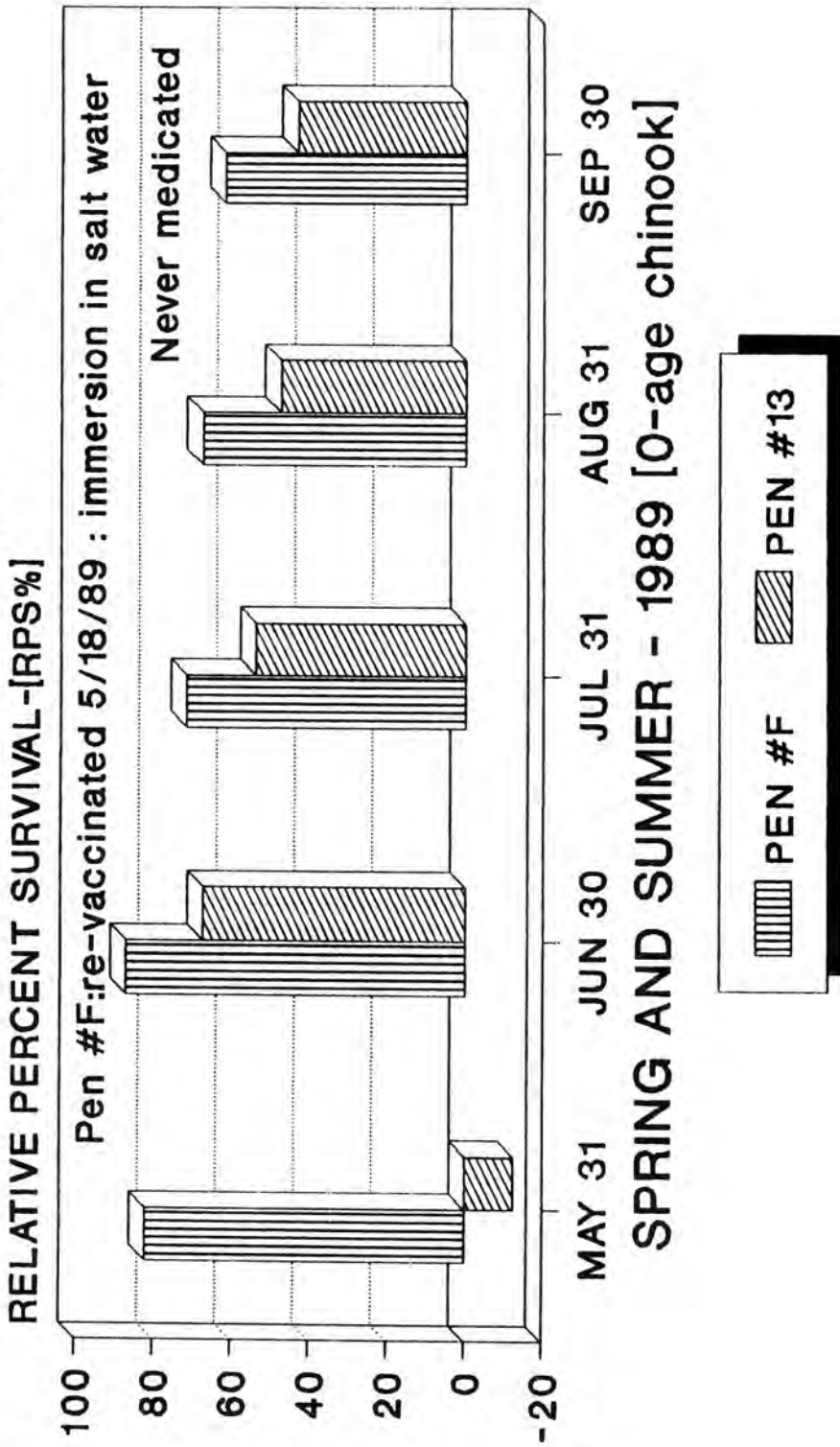
| | PEN NO. | MAY | JUNE | JULY | AUGUST | OCTOBER |
|------------------------------|---------|----------|---------|---------|---------|----------|
| Starting Number of Fish | 13 | 14812 | 14220 | 13614 | 13398 | 13168 |
| Monthly Mortality | 13 | 86 | 506 | 606 | 216 | 230 |
| Cumulative Survivors | 13 | 14726 | 14220 | 13614 | 13398 | 13168 |
| Avg.Wt.(g) at vaccination | 13 | | | | | |
| Total Wt.-Kgs | 13 | | | | | |
| BIOVAX cost at C\$85.50/L | 13 | | | | | |
| Man-hours required | 13 | | | | | |
| Labor cost @ C\$7.00/hour | 13 | | | | | |
| ROMET 30 used - kgs | 13 | | 0.74 | 0.12 | | |
| ROMET 30 cost @ C\$103/kg | 13 | | \$76.22 | \$12.36 | | |
| Tribrissen used - kgs | 13 | | | 0.12 | | |
| Tribrissen cost @ C\$159/kg | 13 | | | \$19.08 | | |
| Tribrissen feed used - kg | 13 | | | 43.20 | 59.66 | |
| Tribr. feed cost @ C\$522/mt | 13 | | | \$22.55 | \$31.14 | |
| Monthly cost of medication | 13 | | \$76.22 | \$53.99 | \$31.14 | |
| Total cost of medication | 13 | | | | | \$161.35 |
| Cost of drugs+vaccination | 13 | \$161.35 | | | | |
| Avg. wt. at month end (g) | 13 | 20.7 | | | 139 | 350 |
| Biomass at month end (kg) | 13 | 304.8 | | | 1862.3 | 4608.8 |
| Cumulat. total cost/kg fish | 13 | | | | \$0.09 | \$0.05 |
| Month end biomass gain (kg) | 13 | | | | 1597.5 | 4304.0 |
| Cost/kg of biomass gain | 13 | | | | \$0.10 | \$0.04 |
| Cumulat.cost/1000 survivors | 13 | | | | \$12.04 | \$12.25 |

The complete report to the U.S.D.A. and the C.M.A. is being edited, and BIOMED, Inc. prepared a BIOMED TECHNICAL BULLETIN last summer on "salt water re-vaccination with BIOVAX". If you'd like a copy of either the report or the "how to do it" bulletin, write, FAX or call BIOMED, INC., 1720-130th Ave. N.E.,

Bellevue, WA. 98005-2203; FAX: (206) 882-2678; phone:(206) 882-0448, and ask for the report: "1989 Hardy SeaFarms/BIOMED, Inc. field trials:re-vaccination of chinook salmon in marine net-pens with BIOVAX vaccines to prevent vibriosis", and BIOMED TECHNICAL BULLETIN No. 1.

HARDY SEAFARMS-VIBRIOSIS RE-VACCINATION

Relative Percent Survival-RPS: Chinook
 Research pen #F and medicated pen #13
 RPS-% is compared against Production Control Pen #11



BIOVAX re-vaccination field trials
 Pen #11 [control]: never medicated
 single, primary fresh-water vaccination

Pen #13: single, primary fresh-water vaccination
 medicated as needed for production

A SUMMARY OF INFECTIOUS HEMATOPOIETIC NECROSIS VIRUS TRANSMISSION STUDIES IN OREGON

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Studies conducted in Oregon concerning the transmission of infectious hematopoietic necrosis virus (IHNV) in hatchery reared salmonids can be summarized under three major topic headings. These are: 1) vertical and covert transmission of IHNV; 2) the effect of water flow and formalin treatments on the prevalence of IHNV in adults salmonids; and 3) other factors that may influence the IHNV carrier rate in infected broodstocks.

Vertical and Covert Transmission Studies

Vertical transmission studies have examined the potential of progeny from IHNV carrier adults to contract the disease via infected or contaminated gametes. Production trials have been conducted throughout the state where progeny from carrier and virus-free parents were kept segregated and monitored for infectious hematopoietic necrosis (IHNV). To date over 9 million fish from IHNV positive parents, representing two different anadromous species and four different

stocks, have been reared with no evidence of virus (Table 1). In most cases, eggs were either water-hardened or surface disinfected with iodophor and in all tests, egg incubation and early rearing of fish was done in virus-free water.

Covert transmission studies have examined the potential of progeny from IHNV positive parents to return as carriers of the virus. For three years at Elk River Hatchery and in 1986 at Bonneville and Wallowa hatcheries, progeny from carrier and virus-free parents were differentially marked prior to release. These fish are then monitored as returning adults for IHNV. In 1987 Elk River, jack salmon (two year adults) from the 1985 brood year returned, and in 1988, jacks for the 1986 brood year returned along with three year old adults from 1985 releases. No virus was detected in any of these fish whether they were from carrier or virus-free parents (Table 2). Marked adults at these facilities will continue to be evaluated for the next five years.

Table 1. Production trials of rearing progeny from adult salmonids infected with infectious hematopoietic necrosis virus (IHNV).

| Facility | Species | Year | IHNV Carrier Rate | | Resulting Progeny | | IHNV Tests ^a |
|-------------|-----------|------|-------------------|------|-------------------|----------|-------------------------|
| | | | Female | Male | Positive | Negative | |
| Bonneville | Chinook | 1986 | 63% | 45% | 3.5 mil. | 0.5 mil. | NEV |
| | | 1987 | 52% | 42% | 4.7 mil. | 1.2 mil. | NEV |
| Round Butte | Steelhead | 1984 | 60% | 21% | 64,000 | 64,000 | NEV |
| | | 1985 | 71% | 16% | 100,000 | 100,000 | NEV |
| | | 1988 | 71% | 12% | 60,000 | 19,000 | NEV |
| Elk River | Chinook | 1985 | | 10% | 70,000 | 50,000 | NEV |
| | | 1986 | | 37% | 75,000 | 50,000 | NEV |
| | | 1987 | | 7% | 75,000 | 50,000 | NEV |
| Irrigon H. | Steelhead | 1986 | 22% | 48% | 0.5 mil. | 1.1 mil. | NEV |

^a No evidence of virus.

Table 2. Prevalence of infectious hematopoietic necrosis virus (IHNV) in returning adults at Elk River Hatchery to determine if covert transmission of IHNV occurs.

1987-88 Results of Marked Fall Chinook Jacks: 1985 Brood Year

| IHNV Status of Parents | Mark | Number Sampled | Proportion of Sample Pools IHNV(+) | | | Result ^a |
|------------------------|------|----------------|------------------------------------|--------|------|---------------------|
| | | | Milt | Spleen | Gill | |
| Negative | LV | 16 | 0/16 | 0/16 | 0/16 | NEV |
| Positive | RV | 12 | 0/12 | 0/12 | 0/12 | NEV |

1988-89 Results of Marked Fall Chinook Jacks: 1986 Brood Year

| IHNV Status of Parents | Mark | Number Sampled | Proportion of Sample Pools IHNV(+) | | | Result |
|------------------------|------|----------------|------------------------------------|--------|------|--------|
| | | | Milt | Spleen | Gill | |
| Negative | LV | 35 | 0/35 | 0/35 | 0/35 | NEV |
| Positive | RV | 20 | 0/20 | 0/20 | 0/20 | NEV |

1988-89 Results of Marked Fall Chinook Adults: 1985 Brood Year

Males

| IHNV Status of Parents | Mark | Number Sampled | Proportion of Sample Pools IHNV(+) | | | Result |
|------------------------|------|----------------|------------------------------------|--------|------|--------|
| | | | Milt | Spleen | Gill | |
| Negative | LV | 25 | 0/25 | 0/25 | 0/25 | NEV |
| Positive | RV | 22 | 0/22 | 0/22 | 0/22 | NEV |

Females

| IHNV Status of Parents | Mark | Number Sampled | Proportion of Sample Pools IHNV(+) | | | Result |
|------------------------|------|----------------|------------------------------------|--------|------|--------|
| | | | Ovarian Fluid | Spleen | Gill | |
| Negative | LV | 12 | 0/12 | 0/12 | 0/12 | NEV |
| Positive | RV | 2 | 0/2 | 0/2 | 0/2 | NEV |

^a

No evidence of virus.

Effect of Water Flow and Formalin Treatment on the Prevalence of IHNV in Adults

In spawning populations of anadromous salmonids in which some individuals are infected with IHNV, the prevalence of infected adults usually increases and may approach 100% as the spawning run progresses. Prior to sexual maturation, adult salmonids are often held at high densities and in low flows at hatcheries. This could create a situation in which a few fish harboring IHNV might infect the remaining population by horizontal

transmission of waterborne virus. Therefore, horizontal transmission may be an important mechanism for dissemination of the virus and among adult salmonid populations.

The rate of water flow into adult holding ponds did not appear to affect the prevalence of IHNV carriers between replicate groups of approximately 300 chinook salmon (*Oncorhynchus tshawytscha*) held at rates which differed by 600 gal/min (Table 3). However, lower carrier frequencies were attributed to formal treatments. In one study, replicate groups

of adult steelhead (*O. mykiss*) were held under similar conditions but the cater of one group received 167 ppm formalin for 80 minutes, 5 days per week and the second was left essentially untreated (control). The prevalence of IHNV carriers among female steelhead that received the formalin treatments was 18% lower than in those left

untreated. No differences were seen in steelhead males (Table 4). When this study was repeated on a later migrating group from the same stock of fish, no difference was seen in the females; however, males that received the formalin treatments had a 33% lower carrier rate than males left untreated (Table 5).

Table 3. The prevalence of infectious hematopoietic necrosis virus (IHNV) in adult chinook salmon (*Oncorhynchus tshawytscha*) in ponds at Round Butte Hatchery receiving two different water flow rates.

| Rate of Water Flow (gal/min) | Proportion of Adults | | IHNV (+) Combined |
|------------------------------|----------------------|----------------|-------------------|
| | Females | Males | |
| 900 | 56/81 (69%) | 11/23 (48%) | 67/104 (64%) |
| 300 | 50/72 (69%) | 8/23 (35%) | 58/95 (61%) |

Table 4. The effect of formalin treatments on infectious hematopoietic necrosis virus (IHNV) in steelhead (*Oncorhynchus mykiss*) adults at Round Butte Hatchery.

| Treatment | Proportion of Adults | | IHNV (+) Combined |
|-----------------------|----------------------|-----------------|-------------------|
| | Females | Males | |
| ^a Formalin | 53/82 (65%) | 21/104 (20%) | 74/186 (40%) |
| No Formalin | 57/70 (83%) | 3/72 (15%) | 60/142 (42%) |

^a The formalin-treated group received 167ppm formalin for 80 min, 5d per week for three months.

Table 5. The effect of formalin treatments on infectious hematopoietic necrosis virus (IHNV) in steelhead (*Oncorhynchus mykiss*) adults at Round Butte Hatchery.

| Treatment | Proportion of Adults | | IHNV (+) Combined |
|-----------------------|----------------------|----------------|-------------------|
| | Females | Males | |
| ^a Formalin | 35/38 (97%) | 6/17 (35%) | 41/55 (75%) |
| No Formalin | 38/40 (95%) | 23/34 (68%) | 61/74 (82%) |

^a The formalin-treated group received 167ppm formalin for 80 min, 5d per week for one month.

In these tests, some formalin-treated adults had a lower IHNV carrier rate than untreated fish. In vitro concentrations of formalin ranging from 100 to 1000 ppm reduced waterborne IHNV concentrations by 53 to 92% in 1 hour, respectively; whereas, 1 ppm concentrations of malachite green reduced IHNV concentration by only 20% in the same time period. It is possible that the virucidal activity of formalin functions to keep the concentration of virus in the water below a threshold needed for efficient horizontal transmission to occur.

Other Factors that Might Influence the IHNV Carrier Rate

Evidence suggests that adult salmonids become infected with IHNV. Therefore, the hatchery practices involved with the handling of these adults may influence their subsequent infection. This is particularly important when the hatchery design is conducive to horizontal transmission between adult salmon and/or steelhead. Critical evaluation of facility design can identify potential areas where horizontal transmission could be occurring. Areas identified included the routing of adult holding pond effluent into the fish ladder, the piping of drainage from egg taking activity (including blood, ovarian fluid, and egg rinses) back into the adult holding pond or fish ladder, and the anesthetic bath water into which all adults are placed prior to sorting and spawning. Significantly, IHNV has been detected in water samples taken from anesthetic baths at two facilities. Concentration of virus detected ranged from 20,000 to 60,000 virus/L. Potential sources of virus in the

anesthetic water include eggs and ovarian fluid expressed from fish being anesthetized and mucus sloughed from the external surface of adult. Mucus collected from the external surface of infected adult and juvenile salmonids can contain IHNV in concentrations ranging from 10 to 1,000,000 virus/mL and could serve as a significant source of virus. This study identified potential sources where viral contamination could occur and led to the development of management strategies that could minimize the transmission of IHNV between adult salmonids.

Acknowledgements

The successful completion of these studies has been the result of tremendous cooperation throughout the Oregon Department of Fish and Wildlife. Significant contributions include Jerry Russum and his crew at Elk River Hatchery and the Elk River Research Section, Ray Hill and his crew at Irrigon Hatchery, John Isley and his crew at Wallow Hatchery, Trent Stickell and his crew at Bonneville Hatchery, and Randy Robart and Bill Nyara and their respective crews at Round Butte Hatchery. The agency's Fish Pathology Section, particularly Warren Groberg and John Kaufman, were key contributors to these study results.

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THE EFFECTS OF IODINATED AND OZONATED WATER SUPPLIES ON STEELHEAD TROUT SUSCEPTIBILITY TO THE IHN VIRUS

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ABSTRACT

A study was conducted to determine the effects of iodinated and ozonated water supplies on the susceptibility of steelhead trout (*Oncorhynchus mykiss*) to infectious hematopoietic necrosis virus (IHN). Eyed steelhead eggs from IHN negative adults were pooled, subdivided, disinfected with iodine, and hatched in egg jars. Mortalities were recorded for the 65 days following egg hatching. While IHN was confirmed in all iodinated (0.26 mg/l) tanks, chronic iodine toxicity may have contributed to the 80.0 percent average loss. Fish reared in ozonated water lost 6.9 percent of their populations, insignificantly different from control losses. Special hatching and early rearing conditions for experimental fish may have contributed to control losses of only 15.1 percent, compared with 75.5 percent losses experienced by non-experimental steelhead reared in raw water this production year.

INTRODUCTION

Dworshak National Fish Hatchery (NFH) is located at the confluence of the North Fork of the Clearwater River and the main stem of the Clearwater River in north central Idaho. The hatchery water supply is the North Fork of the Clearwater River.

Beginning with steelhead trout (*Oncorhynchus mykiss*, formerly *Salmo gairdneri*) from Brood Year 1982, Dworshak NFH has experienced extensive losses to Infectious Hematopoietic Necrosis Virus (IHN). Various techniques have been tried over the past eight years involving egg selection, incubation, diet, cultural procedures, and water supply alteration to analyze and affect susceptibility to IHN in the various brood years.

Culling eggs from IHN positive adults and irregular attempts at inoculating non-infected fish with fish or water from infected rearing units has had no apparent effect to date. Consistent protection from the virus has only been attained with the use of an ozonated (pathogen-free) single-pass water supply.

Dave Owsley, environmental engineer at Dworshak, learned of promising results Dr. Bob Busch, Director of Rangens Laboratory in Buhl, Idaho, was having rearing rainbow trout (*Oncorhynchus gairdneri*) in water to which iodine was added. At 0.3 to 0.5 mg/l iodine, the rainbow trout (RBT) experienced significant reductions in losses to bacterial gill disease. Confirmation of the viricidal effect of these levels of iodine exposure by Dr. Jim Winton of the Seattle National Fisheries Research Center gave the impetus to a pilot study.

In March and April of 1989, initial testing was conducted of a new iodine delivery system installed at the hatchery. It was demonstrated that a consistent iodine concentration could be maintained and a satisfactory survival of the fish reached at 0.26 mg/l iodine. Fish lethargy and mortality increased over 0.50 mg/l using spring chinook salmon (*Oncorhynchus tshawytscha*) fingerlings. At 0.65 mg/l all fish died. These results confirmed the feasibility of conducting a test on steelhead trout (STT) at Dworshak. An experiment was designed for comparing the effect on IHN susceptibility of rearing Brood Year 1989 steelhead on 1) untreated, raw water (control), 2) iodinated water, and 3) ozonated water.

MATERIALS AND METHODS

The initial experimental design was to raise approximately 25,000 fingerlings in each experimental tank, with four

replicates of each treatment. The different treatment tanks were grouped into contiguous blocks because of plumbing restrictions. Randomization of treatments was, therefore, not possible. The four tanks receiving ozonated water were Tanks 17, 18, 19, and 20, located in A-bank of Dworshak's nursery building. The four tanks receiving iodinated water were Tanks 21, 22, 23, and 24, also located in A-bank. Four tanks receiving raw (untreated) water were Tanks 33, 34, 35, and 36, in B-bank, adjacent to the ozonated experimental tanks. Tank dimensions were 3' wide by 16' length by 2' water depth, a capacity of 96 cubic feet. Ozonated and raw water groups were in concrete tanks and the iodinated groups were in fiberglass tanks.

The ozonated system, in operation for several years at Dworshak, consisted of two ozone generators and a contactor. The ozone was removed by packed columns. No measurable ozone residual could be detected in the water when it entered the experimental tanks. The iodination system consisted of an iodine saturator, a chemical metering pump, a self-powered flow meter, and a pump controller. The iodine concentration, measured and recorded daily, average 0.26 mg/l over the course of the study. Raw water pumped from the North Fork of the Clearwater River was the water supply for the control tanks.

The study began on June 2, 1989 when 319,000 eyed steelhead trout eggs from Dworshak NFH's tenth egg take were prepared for placement in egg jars. These eggs, taken on April 18, were from returning STT adults which had tested negative for IHNV. Eggs were pooled, thoroughly mixed and then evenly divided among the 12 treatment tanks. This resulted in each replicate receiving 26,600 eggs. The individual egg lots were placed into egg jars containing 100 mg/l iodine solutions to which sodium bicarbonate had been added for buffering; Dworshak's water, less than neutral pH, requires buffering when disinfecting with iodine to prevent too low a pH. The water supply to each jar was turned on after disinfecting the eggs for 10 minutes. Flows were adjusted to approximately five gallons per minute to suspend and gently roll the eggs in the water column.

The Dworshak hatching jar consists of a 6-inch diameter pipe (24-inches high), inserted into a 12-inch pipe of lesser height. The flow is forced down in the 12-inch pipe and up in the 6-inch pipe. Eggs are placed in the smaller pipe. The hydraulics allow for an even flow pattern through hatching and swim-up (Figure 1). The hatchery uses 2-3 gallons per minute (gpm) on green eggs, 5 gpm when eyed, and 4 gpm at hatching time.

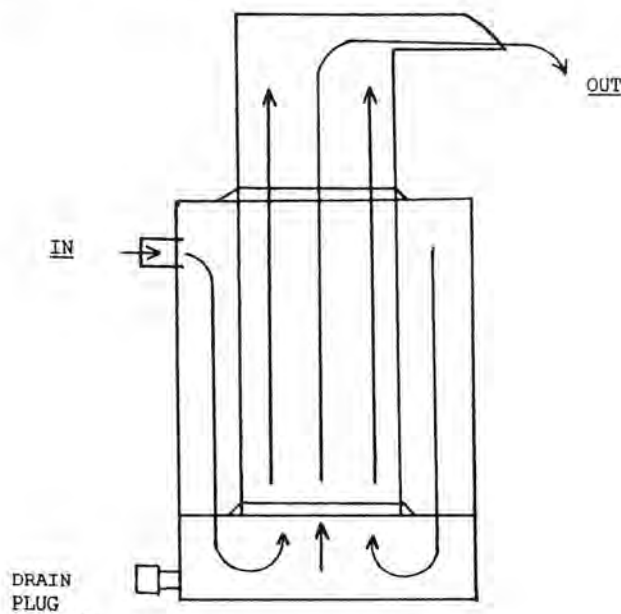


FIGURE 1. Dworshak Hatching Jar
Downward Flow in 12-inch column
Upward Flow in 6-inch column

Hatching began on June 7 and was completed by June 11. Over 75 percent of the fry had completed swim-up by June 23 and more than 50 percent had already left the jars and were in the rearing tanks. On this same date the remaining fry were emptied into the tanks and feeding begun. Hatching jars were removed and water flows increased to 25 gpm at this time.

Water temperature on the day hatching was complete (June 11) was 51.0 F. Over the 65-day experiment the temperature averaged 52.3 F. and ranged from 50.0 F to 55.0 F. Water pH was approximately 6.3 and hardness averaged 10 mg/l.

Feed used during the test period was Oregon Moist Pellet starter mash trout diet from Moore-Clark Company, Inc., LaConner, Washington (product use does not constitute product endorsement by the U.S. Fish and Wildlife Service). Steelhead fingerlings were fed "by eye" eight times per day for the duration of the study, or approximately five percent of body weight daily. Mortalities were removed and tanks were cleaned daily. Each tank had its own mortality net and cleaning brush and care was taken to avoid cross-contamination at all times.

Examination for the presence of the IHN virus in steelhead fingerlings were performed by the Dworshak Fish Health Center (FHC), using bioassay tissue culture.

A one-way analysis of variance using Duncan's Multiple Range Test was used to test for significant difference between cumulative mortalities of the two treatment groups and the control. Alpha was 0.05, and the mortality data was transformed into arcsine proportions. We would like to thank Greg Pratschner, at Leavenworth National Fish Hatchery, for performing the statistical analysis.

The study, terminated after 65 days on August 14, showed two of the four iodine tanks had already lost over 75 percent of their populations to a possible iodine toxicity problem and later to confirmed IHN virus disease.

RESULTS AND DISCUSSION

As shown in Table 1, steelhead fingerlings reared in control conditions in raw water lost an average 15.1 percent of their populations during the study. Only one of the four control tanks actually experienced a confirmed outbreak of IHN. The 5.3 to 8.2 percent losses in the three tanks which remained "clean" were considered normal rearing mortality. In fact, the averaged mortality curve for these tanks closely approximated that of the four tanks reared on ozonated water of which none showed sign of a viral outbreak.

Table 1. Brood Year 1989 steelhead losses to IHNV comparing raw, iodinated and ozonated water supply effects.

| Treatment | Replicates (Percent Losses) | | | | Start No. | Total Morts | Cumul. Morts (%) |
|---------------|-----------------------------|------|-------|------|-----------|-------------|------------------|
| | 1 | 2 | 3 | 4 | | | |
| Control (Raw) | 8.2 | 7.0 | 39.8 | 5.3 | 103,671 | 15,631 | 15.1 |
| Ozone | 7.2 | 6.9 | 7.1 | 6.5 | 103,784 | 7,155 | 6.9 |
| Iodine | 86.1 | 64.7 | 100.0 | 61.9 | 101,981 | 81,562 | 80.0 * |

* Significantly different from control losses at the 0.05 level.

Fish on ozone suffered an average 6.9 percent loss during the study. None of these losses, as mentioned, were demonstrated attributable to the IHN virus. This result confirms earlier observations that provision of a virus-free water supply with IHN-negative eggs can significantly reduce IHNV losses. What is intriguing about this study is that there was no significant difference in losses between ozone-reared fish and control fish. As mentioned above, only one of four control tanks broke with the disease.

One might conclude that the virus level in Dworshak's raw water supply was abnormally low to have both raw and ozone water supplies yield such similar low mortalities. However, during the same period this study was conducted, 2.6

million steelhead fry from Takes 10, 11, and 12 were being reared in the same raw water supply as the control fish. These fish had all been incubated, hatched, and partially buttoned-up in trays of Heath incubator stacks on raw water. Eggs had not been disinfected with iodine at eye-up as had all experimental eggs. As shown in Table 2, losses among these fish during the first 65 days after hatching averaged 75.5 percent. The only differences between this group and the control were that control fish were disinfected at eye-up and hatched in up-welling hatching jars. The jars removed shell debris and provided uniform, clean rearing conditions. These circumstances could evidently be enough to reduce IHN losses in control fish to approximately the same as those in ozonated water.

Table 2. Brood Year 1989 steelhead losses to IHNV in non-experimental tanks during the initial 65-day rearing period.

| Egg Take | IHN Status of Adult STT | Rearing Water | Start No. | Total Morts | Cumul. Morts (%) |
|---------------------|-------------------------|---------------|-----------|-------------|------------------|
| 10 | Positive | Raw | 266,077 | 199,277 | 74.9 |
| 11 | Positive | Raw | 843,346 | 624,822 | 74.1 |
| 12 | Negative | Raw | 763,048 | 459,334 | 60.2 |
| 12 | Positive | Raw | 728,188 | 679,375 | 93.3 |
| Totals /Ave for Raw | | | 2,600,659 | 1,962,808 | 75.5 |
| 11 | Negative | Ozone | 190,650 | 53,739 | 28.2 |

Note: Statistical comparisons are not made between these data and data from experimental tanks. Values in this table are from concurrent production program and presented for discussion only.

One control tank lost only 39.8 percent of its population, compared with losses of 60 to 100 percent in normal production tanks breaking with IHNV. Special handling procedures may have an effect not only on whether a tank becomes infected with the disease but also on the severity of the outbreak. It may be that sac fry have a greater chance of becoming infected with the virus when eggs are not disinfected

with iodine at eye-up and when hatching occurs in incubator trays having less uniform circulation. Steelhead fry hatching under these conditions may be more susceptible to the virus than if contacting the disease at a larger size when, presumably, their immune-response systems would provide greater protection. The outer shells of all experimental eggs were disinfected at eye-up before hatching

and all debris expelled from the jar by the water flow at hatching. This procedural change could likely have prevented the emerging sac fry from being exposed to shell-borne virus and limiting viral exposure only to that in the entering water supply.

The proportion of IHNV losses attributable to hatching and early rearing conditions remain to be demonstrated. This also holds true for losses attributable to the water supply. This point is demonstrated by the 28.2 percent average losses experienced by Take 11, a negative IHN-parentage steelhead group reared in ozonated water at the same time this study was taking place (Table 2). These fish were not disinfected at egg eye-up and were partially buttoned-up in Heath stack trays on raw water prior to being transferred to the treated ozonated water supply. (NOTE: normally if fish are to be reared in an ozone treated water supply, eggs would have incubated and hatched on similar water conditions). While 65-day losses ranged from 3.7 to 63.2 percent, only one tank lost a majority of its population. Fish in the high mortality group evidently acquired the disease in incubator stack trays similar to the groups in raw water which lost 75.5 percent of their populations. It is possible that extensive losses occur only when early infection with virus in incubator trays is combined with subsequent rearing in virus-laden water. Early infection in the incubation and hatching stages followed by virus-free rearing conditions may still result in losses, however, these losses may be considerably reduced.

This may also explain the low losses in the experimental control fish. These fish did not contact the virus immediately after hatching because of improved hatching and early-rearing conditions (disinfected eggs and jar hatching). One tank broke with IHN and lost only 39 percent of its population possibly due to the larger size fish at time of infection.

Hatching procedures dramatically reduced losses in experimental control fish. However, these steps were not enough to prevent IHNV in the fish reared in tanks with iodine treated water. The four iodine tanks all experienced confirmed

virus outbreaks with an average loss of 80 percent (Table 1). Iodine at 0.26 mg/l in soft, acidic water may not be an effective viricide against this particular strain of IHNV. The conclusion may be drawn that iodine actually made the steelhead fingerlings susceptible to the virus when considering the low losses experienced by the control fish. It should be noted that in a memorandum of August 24, 1989, to the Dworshak FHC Dave Owsley stated that at a pH of 8, iodine is predominantly (88 percent) in its viricidal form, hypoiodous acid. At Dworshak NFH's pH of 6.3, the elemental (and most bactericidal) form predominates.

Rainbow trout subsequently exposed in a later study to chronic low iodine levels experienced considerable losses. Histological examinations revealed kidney and gill damage which may have caused the problem. Insufficient evidence exists in the steelhead study to ascertain what proportion of iodine-treated fish actually succumbed to chronic iodine toxicity rather than to IHNV.

CONCLUSIONS

The addition of iodine at 0.26 mg/l to Dworshak NFH's low pH water supply did not confer protection from IHNV to the steelhead trout currently being reared. Iodine addition may have made the fish more susceptible to the virus or the level of iodine added in Dworshak's soft, low pH water was toxic to the fish. Further attempts to use iodine at Dworshak NFH will be made if more controlled laboratory testing at the Seattle National Fisheries Research Center indicates its continuation is warranted.

Disinfection of eggs at eye-up and the subsequent hatching and early-rearing of the fry in egg jars instead of incubator trays appear to substantially reduce losses to the IHN virus. These steps will now become standard procedure for all production fish held at Dworshak.

Comparisons of losses to the virus between study and normal production steelhead groups (Brood Year 1989) demonstrate the need to examine further the proportional contribution to IHNV losses by varying disinfection, hatching, and early-rearing

methods. Table 3 illustrates the general experimental design for a study proposed for Brood Year 1990 steelhead. A total of 640,000 steelhead trout eggs will be pooled and then subdivided into eight

groups of variable combinations with four replicates from each group. Hopefully, these numbers of fish and of replicates will be sufficient to evaluate the relative contributions of the variables discussed.

**Table 3. Hatching Protocol IHN Effects Study
Proposal- STT Brood Year 1990.**

| Disinfectant | Hatching | Rearing |
|---------------------|-----------------|----------------|
| Yes | Jar | O ₃ |
| Yes | Jar | Raw |
| Yes | Inc | O ₃ |
| Yes | Inc | Raw |
| No | Jar | O ₃ |
| No | Jar | Raw |
| No | Inc | O ₃ |
| No | Inc | Raw |

A SIMPLE ECONOMICAL STATIC MIXER FOR CHEMICAL TREATMENT OF EGGS

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Chemical treatment of fish eggs for fungus control is common practice in hatcheries. Aquatic fungi, especially *Saprolegnia parasitica* are present in all hatchery water supplies (Woods, 1974). Fungi tend to establish themselves on dead organic material with dead eggs providing an excellent growth medium. The fungi growing on dead eggs soon expand and can smother adjacent live eggs. In severe cases, the entire group of eggs can become solid masses of fungus.

Picking the dead eggs will control fungus growth. When the eggs are in the "tender stage", this is not possible. Hatcheries dealing with large numbers of eggs experience a manpower and time consumption problem. Therefore, most hatcheries use some type of chemical treatment to control fungus.

In the past, chemical treatments consisted of the "California flush" method, drip bottles, chicken waterers, and chemical pumps. Because of new safety regulations, only the chemical pump method with a closed system is acceptable with the new OSHA regulations.

Although many hatcheries employ use of Heath trays for egg incubation, more emphasis is being directed to single parent incubation. This emphasis is primarily for the control of Infectious Hematopoietic Necrosis (IHN) and Bacterial Kidney Disease (BKD). Buckets and colanders are now used in many facilities for egg incubation.

Regardless of the style of incubation or species being reared, the application of chemicals for treatment has similarities. The chemical must be introduced into a water supply and delivered to eggs.

Most systems consist of a water supply line with multiple discharge points for each

incubator. For Heath trays, chemical treatment can be administered by a pump with separate fixed flow nozzles for each stack of trays. This system works fine with a flow of 5 to 10 gpm per stack. Bucket and colander incubation is not as easy to administer because of the small flows typically of two-thirds, three-fourths, to 1 gpm.

At Dworshak, chemical treatment for bucket and colanders is achieved by injecting the chemical into a single supply line. This was not an effective way of treating because of the pipe length, number of outlets, and laminar flow within the pipe.

A simple mixing device was constructed using aluminum biorings manufactured by Koch Engineering Company. The aluminum rings are 3 1/2 inches in diameter by 3 1/2 inches in length. Three rings were welded together for a total length of 10 1/2 inches. The four inch water supply was flanged so that the mixer could be removed for cleaning.

The mixer was installed above or after the chemical injection point. Using a colored dye and colorimeter, tests were conducted before and after the mixer installation. Tests showed a uniform mix with the static mixer. A non-uniform mix is present without the static mixer.

This simple device that can be built and installed for approximately \$50 is comparable to a \$750 unit that is commercially available. These units can be built from plastic or metal biorings and will improve efficiency of chemical mixing for treatment. The biorings come in various sizes including 5/8 inch, 1 inch, 1 1/2 inch, 2 inch, and 3 1/2 inch to accommodate most water supply lines. The mixers should only be installed where they can be removed periodically for

examination and cleaning. This can be accomplished by incorporating bolt type flanges.

A list of manufacturers that make biorings include: *

1. Koch Engineering Co. Inc.
6880 Orangethorpe Ave. Unit C.
Buena Park, CA 90620
(714) 739-8853
2. Glitsch, Inc.
P.O. Box 226227
Dallas, TX 75226
(214) 631-3841
3. Jaeger Products, Inc.
P.O. Box 1563
Spring, TX 77383
(800) 678-0345
4. Norton Co. Performance Plastics
Box 3660
Akron, OH 44309
(216) 798-9240

* The companies listed above are not to be considered the only manufacturers of biorings, nor are they endorsed by this paper.

This type static mixer offers good mixing and low head loss. The pressure drop or head loss was not measurable in the 4 inch line at Dworshak. It is important that the chemical being used to treat eggs is mixed thoroughly so that each container of eggs receives the same treatment.

Reference

Wood, J.W. 1974. Diseases of Pacific Salmon, their prevention and treatment. State of Washington Department of Fisheries, Hatchery Division, Technical Publication, 2nd Edition.

STRAWBERRY DISEASE: POSSIBLE CAUSES AND METHODS OF CONTROL

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Strawberry Disease (SD) is a nonfatal skin condition of salmonids that is characterized by raised, red-stippled, dry lesions in fish 5/# or larger.

Species that have SD-like lesions include rainbow trout (*Oncorhynchus mykiss*), cutthroat trout (*O. clarki*), and whitefish (*Prosopium williamsoni*). Dermal necrosis in chinook salmon (*O. tshawytscha*) appears very similar to SD due to the focal, dry nature of the lesion, the lack of mortality or behavioral changes, and the lack of associated pathogen.

SD has been reported in Washington, Oregon, California, Nevada, Idaho, Montana, Colorado, British Columbia, and southern France.

There are many differing opinions on the cause of the condition. The most common ideas include an atypical strain *Aeromonas salmonicida*; an allergic, or hypersensitive, response to the feed or environment; a mycoplasma; and adeno-like virus, or a member of the Rickettsiaceae family, such as chlamydia.

My research attempted to pinpoint the exact cause of SD. A bacterial agent was assumed to be the cause due to reportedly successful transmission studies, as well as, response to treatment with oxytetracycline medicated feed.

Sections of the lesion itself were examined and no pathogens were seen in the sections by routine histological stains.

Several bacterial species were cultured from the lesions and the kidney but all were common flora of the fish skin and water. Because of their widespread nature and because they were also isolated at facilities with no history of SD, these bacteria are not considered the likely cause of SD.

However, homogenate made by grinding up SD lesions exhibited cell destruction in repeated passages onto EPC cell cultures. The inoculated cell cultures were examined with transmission electron microscopy and an irregularly shaped, membrane bounded organism was observed in two of four SD homogenates sampled. This organism was not seen in control cell cultures inoculated with homogenate prepared from normal fish skin. The appearance of these organisms is consistent with members of the Rickettsiaceae family.

Kinyoun's acid-fast and Pinkerton-Rickettsiaceae stains were applied to sections of SD lesions and displayed positive red inclusion bodies in a small number of cells. Rickettsial species are acid-fast and the intracellular location of positive red inclusions in the Pinkerton-Rickettsia stain is indicative of a member of the Rickettsiaceae.

This information, together with the organism seen in electron microscopy, the growth on the cell culture but not on artificial media, as well as the difficulty in isolating an agent, are all characteristic of the Rickettsiaceae. Further work to purify the agent and demonstrate lesion development as well as the ability to reisolate the organism from the lesion need to be performed to prove it to be the cause of SD. Serological analysis with antibody to members of the Rickettsiaceae family should be performed on SD inoculated cell cultures and SD lesions to assist in the identification of the agent and further substantiate its association to the condition.

I also performed oxytetracycline trials on fish with SD at the Clear Springs Trout Company Research and Development Laboratory and Snake River Trout Farm in Idaho to determine whether treatments improved the healing rate of SD lesions.

Fish with SD lesions were taken from one raceway for each study and split into two tanks in the wet lab or live boxes in a SD positive raceway. Half of the fish were fed with TM100 and the other half with non-medicated production feed. Eight live boxes were used in the first production facility study and four were utilized in the second run.

Fish were fed medicated feed for 14 days (4 gm/100 pounds of fish/day) and then were placed on non-medicated feed for the duration of the study. The experiment ran for five to six weeks and fish were examined weekly in the lab and every other week in the live boxes for the presence of lesions.

Results were analyzed with the student t test. One of the laboratory runs was found to have a statistically significant difference in the healing rates between the medicated and unmedicated fish. The other laboratory experiment and the two production facility experiments did not demonstrate a statistically significant improvement in healing between the medicated and unmedicated fish.

The ambiguity of these results may be interpreted several ways. There may be a limited effect of oxytetracycline on the causative agent of SD. We may be treating the general health of the fish and not directly effective against SD at all. The causative agent of SD may have developed a resistance to oxytetracycline from repeated use over the years. Effectiveness of treatment needs to be further examined to determine whether medication is beneficial and economical. Recurrence of SD has been observed recently in populations of fish after the initial treatment and healing. This could also support the idea of a developing resistance to oxytetracycline.

Work done by the Nevada Department of Fish and Game in the 70's compared the use of Furazolidone, Terramycin, malachite green, beef liver supplements, and transfer of affected to a different

facility but found none of these methods improved healing over control fish. Romet has also been used with limited success by several commercial trout facilities though no controls were maintained due to limited space. This medication is less desirable to the commercial growers because of the 45 day post treatment withdrawal requirements.

Prevention of SD is not effective at this time as we don't know the cause of the condition. As with most fish diseases, minimizing stress appears to minimize the presence of SD. There is limited benefit in controlling the condition once it is present with oxytetracycline. I strongly encourage controlled studies on split lots of SD fish at each facility before resources are spent on medication.

To summarize these experiments; there is compelling evidence that the causative agent of SD is a member of the Rickettsiaceae family and that treatment of the condition with oxytetracycline is, at best, marginal.

Areas I feel would be productive to investigate include: presence of antibody in the blood sera to a member of the Rickettsiaceae family; development of an autologous antisera for labelled probing of the lesion; purification of the organism present on cell culture and transmission trials to determine if SD lesion development occurs; and further medication trials at a facility with little historical antibiotic use.

I would like to acknowledge my funding sources for this work:

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Clear Springs Trout
Company Silver Cup Feeds
U.S. Trout Farmers Association.

I would also like to express my appreciation to the commercial trout industry, especially in the Hagerman Valley of Idaho, for their unlimited support and cooperation in this project.

**SUPPLEMENTATION STRATEGIES FOR THE PROPOSED
YAKIMA/KLICKITAT PRODUCTION FACILITY**

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Tom Vogel-Bonneville Power Administration**

The purpose of the Yakima/Klickitat Production Project as stated by the Northwest Power Planning Council is to: "...test the assumption that new artificial production can be used to increase harvest and enhance natural production while maintaining genetic resources", with explicit emphasis on experimentation and evaluation to achieve this goal. This emphasis on experimental goals provided impetus for the formation of an Experimental Design Work Group (EDWG) that is comprised of fishery scientists from the cooperating management entities and other groups. EDWG has been meeting biweekly to design experimental, monitoring and evaluation programs for the YKPP. This paper describes the current status of the planned supplementation strategies for the Yakima Basin.

The stocks to be supplemented in the Yakima Basin include spring chinook, summer chinook, fall chinook, coho, sockeye and summer steelhead. Historic run size of all anadromous salmonids in the Yakima system has been estimated to be about 500,000 adults. The current runs of all species range from about 6,000 to 15,000 adults composed of from 4,000 to 10,000 spring chinook, 1,500 to 3,000 steelhead, and 1,000 to 2,000 fall chinook. There are currently no coho, summer chinook or sockeye salmon populations in the Yakima basin. This paper will be concerned mainly with the spring chinook and summer steelhead populations in the Yakima basin.

There are a number of unanswered questions related to the physical and biological resources of the Yakima River that constrain our experimental design task.

For instance, the basic sub-population structure of naturally reproducing species within the basin has not been defined sufficiently to finalize hatchery broodstock and release strategies or genetic monitoring approaches. In addition, fish monitoring capabilities need to be expanded to evaluate the success of the project. These needs are being addressed during the pre-facility design and baseline data collection phase. The proposed hatchery is scheduled for completion in 1994-5 so we have about 5 years to complete the baseline data collection and finalize experimental designs.

The Yakima/Klickitat Production Project is intended to provide new knowledge regarding the application of supplementation techniques. Supplementation acts to increase natural production through improved juvenile survival and increased production capacity, thus shifting the productivity of a system from its current level upward to a higher potential productivity level. Consequently the experimental goal of the project is to design and conduct experiments to resolve critical uncertainties regarding supplementation with a high degree of certainty.

Supplementation of natural populations occurs at the spawner stage. The natural spawning population is supplemented by returning adults from hatchery reared fish. In order to return hatchery fish to the desired spawning area they will be acclimated and released as smolts in or near those areas. The intent is to minimize interactions between outmigrant hatchery fish and natural populations.

In order to achieve supplementation success it will be necessary; 1) for hatchery fish to survive and return to the target spawning area, 2) for the supplemented population (mixture of hatchery fish and natural origin fish) to reproduce successfully, 3) that the fitness of the supplemented population is sustained in the long term, and 4) that the effects on other populations and species be understood and considered in the management process.

Genetic diversity is an important concern for certain species in the Yakima. This concern will be addressed in part by attempting to maintain large natural spawning populations and to supplement them with hatchery fish as described in the following sections.

Hatchery Operation Standards and Assumptions

The intent of identifying hatchery production guidelines for the Y/KPP is to insure that basic goals/objectives of the project are adhered to, i.e. that hatchery practices are consistent with the experimental plan. Since the experimental plan is still evolving, the operational standards and assumptions presented herein are continually being evaluated and reassessed as new information becomes available.

Hatchery practices (e.g. rearing density, diet, general fish culture techniques) have not been considered the most critical uncertainties within the Y/KPP. Because of this and the fact that experimental opportunities will be limited during project implementation, hatchery practices generally will be managed as experimental "constants". A number of questions have been identified within the hatchery production cycle, however, that need to be evaluated with respect to the intent of minimizing potential behavioral or genetic impacts from the hatchery program. These information needs will be addressed through pre-facility studies and planning efforts in order to formulate a final set of hatchery guidelines.

Current guidelines and information needs are outlined in three areas: (1) broodstock collection and mating; (2) rearing; and (3)

release/acclimation.

Broodstock Collection and Mating

The broodstock aspects of the hatchery program may be the most central to minimizing adverse genetic effects on natural populations. Several concepts have been adopted:

1. Unique sub-populations that exist within the sub-basins will be individually cultured within the hatchery program.
2. Broodstock will be collected from progeny of natural spawners (all hatchery fish will be marked to enable this) from these sub-populations.
3. A maximum of 15-20% of any natural-origin adult population will be collected for broodstock (see Refined Goals report) to maintain a large, naturally reproducing gene pool.
4. Hatchery spawning groups will consist of 200 adults with equal numbers of each sex, using fertilization techniques to "equalize" contribution potential of each male.
5. Representative portions of each run will be taken as gamete source in relationship to run timing and age/size composition; use of jacks will consider their potential to actually contribute in a natural spawning situation.
6. Random mating will be used.
7. Routine fish health screening at time of spawning.
8. Detailed records will be kept on all broodstock taken, e.g. disease incidence, return time, age/size/sex, genetic profiles etc. - as a basis for racial/sub-stock separation of broodstock for mating and for comparing hatchery and naturally spawning stock characteristics annually and over time.

Rearing

The general rearing objective is to produce a smolt ready to migrate during or shortly after natural smolt emigration time, at a size (and age) which is as closely reflective of naturally reared smolts as possible. Emigration readiness is obviously important to avoid competitive or predacious interactions with naturally rearing juveniles, while size/age consistency is important for maintaining the basic maturation, migration and life history characteristics of naturally spawning stocks. This objective has already received much discussion with respect to steelhead, as the desired intent for mimicking natural smolt ages likely would have profound operations, performance and experimental consequences. Yakima steelhead are currently programmed for 100% yearling release.

Other rearing guidelines include:

1. Fish selected for experimental treatment/controls and subsequent planting should be selected from as many spawning adults as possible, by taking equal portions of egg groups rather than allocating total egg lots which represent a limited number of females.
2. Uniform rearing conditions will be maintained amongst treatment/ control groups;
3. Detailed records of disease incidence, growth, smoltification, etc. will be maintained.
4. Densities will be lower than those typically used in the region's hatchery programs.

The facilities and procedures for juvenile rearing in the main hatchery will be developed to allow each of the replicate treatment and control groups to be treated as similarly as possible. This will include having the same water source, feed rates, densities, etc. Many of these requirements have been factored into the facility designs by the engineers.

Main Hatchery Facilities

The main facility layout for the Yakima Basin will include three main hatchery sites to be located at (1) Cle Elum on the Upper Yakima River, and (2) Nelson Springs and (3) Oak Flats on the Naches River. The following section will describe the various species/stocks that are slated for supplementation and where the adult holding, egg take and incubation, and rearing will occur.

Spring Chinook

The Cle Elum site will be used to hold spring chinook adults taken from Roza dam as brood stock for the upper Yakima genetic sub-stock (this may need to be revised to sub-stock as results from genetic studies emerge). The adult spring chinook will be spawned at Cle Elum, eggs will be incubated and juveniles will be reared to produce 1,150,000 fish at release at 15/lb. The fish will be released through 15 acclimation facilities as discussed in following section on acclimation facilities.

The site at Cle Elum will also be used to finish incubation and rear the spring chinook for the Naches River as discussed in the following paragraph.

The Naches spring chinook adults will be taken at Cowiche diversion dam and spawned at Oak Flats. The eggs will be incubated to the eyed stage at Oak Flats then transferred to the Cle Elum main facility for final incubation and rearing. These spring chinook will then be transferred back to the Oak Flats main facility for final rearing and the resultant 450,000 pre-smolts will be transferred to the 6 acclimation sites on the Naches system for release back to the habitat.

Summer Steelhead

The summer steelhead adults will be collected at Cowiche Diversion dam and spawned at the Nelson Springs main facility. The egg incubation and early rearing will occur at Nelson Springs. The final rearing will be split between the Nelson Springs and Oak Flats sites with 50% of the juveniles at each main facility.

The resultant 400,000 pre-smolts will then be transported to 6 acclimation facilities on the Naches system (200,000 fish), 3 acclimation facilities on the Toppenish system (100,000 fish), and 3 acclimation facilities tentatively scheduled for interaction research experiments on the upper Yakima (100,000 fish).

Acclimation and Release

Releases from acclimation sites will be the standard hatchery protocol in order to maximize survival and meet project objectives of hatchery fish successfully contributing to natural spawning escapement. The length of acclimation will be an important experimental variable for some locations. The standard acclimation time has not been defined yet although, at a minimum, will have to allow a recovery period from physiological stress of transportation and provide for an adequate level of imprinting. Other guidelines include:

1. Acclimation "pond" water supply should tap ambient source since an important purpose of acclimation is to eliminate immediate physiological stress and resultant mortality at release caused by differences in the rearing and release environment.
2. Volitional releases will be the preferred release protocol.

Questions regarding acclimation and release include:

1. What is the minimum acclimation time?
2. What affect will acclimation siting and length have on return spawner distribution and straying?
3. What impact will volitional releases have on experimental quality control?

The acclimation facilities for spring chinook and steelhead trout are also being designed to standardize the treatment of

final rearing of the release groups. The acclimation facilities have been standardized as 9,000 ft³ ponds with a density factor of 1.1 (1/2 that of raceways), with a flow of 525 gpm/pond or 1.2 cfs/pond. Each pond will be used to finish rearing and acclimate 75,000 spring chinook smolts at 15/lb or 25,000 to 33,000 steelhead smolts at 7/lb. The ponds will be earthen structures with cobble bottom to simulate natural conditions as much as possible. The ponds will need to be totally drained and may be gravity fed or require pumping in some situations.

There will be three or four clusters of three acclimation sites with the potential for two ponds/site located on the Upper Yakima River. In the initial production stages of the project there will only be one pond constructed per site. This pond will be for acclimation and release of spring chinook. The option of constructing a second pond at each site will be used for the acclimation and release of steelhead trout if that is decided to be the study design after collection of the baseline data on resident trout/steelhead. The current potential locations of these clusters are at the Ellensburg Town Dam, near the main hatchery facility at Cle Elum, and just downstream of the Easton Dam site. There is also the potential for a cluster of sites at the Roza Diversion Dam, but this is a lower priority site due to the limited current natural production in this area.

The tributary locations in the upper Yakima will be located in the Teanaway River (one mainstem site, two sites in the North Fork, and one potential site in the Middle Fork, dependant on the building of a storage reservoir on this fork), on the Cle Elum River, and in the section of the Upper river between Easton and Kechelus dams (this section was designated as a tributary rather than as a mainstem section).

The Naches system will have acclimation sites located on the Bumping River, Little Naches River, Rattlesnake Creek, and the Cowichee Creek. These sites will be the same as those described for the upper Yakima with ponds at each site for 75,000 spring chinook and 33,000 steelhead smolts.

A maximum of three sites will also be developed on the Toppenish Creek system for steelhead releases.

The survival for each of the release groups will be monitored as the smolts migrate out of the Yakima Basin at the Juvenile Evaluation Facility at Prosser Dam and again as the adults return to the basin through the ladders at many of the irrigation diversion dams. Additional

monitoring needs will be developed to determine reproductive success and long term fitness of the supplemented stocks through time.

It is believed that the approach taken in this large scale study of supplementation on the Yakima River will provide valuable information on the best methods of using hatchery fish to supplement natural salmonid populations.

THE SNAKE RIVER FALL CHINOOK SALMON EGG BANK PROGRAM: THE FINAL CHAPTER

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OVERVIEW

The Snake River Fall Chinook Egg Bank Program was established in 1976 with two goals: (1) provide an interim adult holding and juvenile rearing program for Snake River stock fall chinook until the Lyons Ferry Fish Hatchery (FH) could be constructed, and (2) maintain the genetic integrity of this stock during this interim period. The Egg Bank Program continued through the initial years of Lyons Ferry FH operation to assist in its broodstock building process.

BACKGROUND

Legislation under the Water Resources Development Act of 1976 provided hatchery compensation for downstream passage mortality and loss of spawning habitat caused by construction and operation of the four lower Snake River hydropower projects (Figure 1). The compensation measures included provision for a fall chinook salmon intended to return 18,300 adults to the project area. The salmon stock generally accepted as appropriate for the project area was the bright fall chinook of Snake River origin.

It was recognized early in the hatchery site selection and planning stages following the enabling legislation that Snake River fall chinook stocks were in a critically depressed status and, in fact, were under consideration for classification as an endangered species (Utter and Ebel 1981; Figure 2). Fish and wildlife agency biologists generally agreed that the Snake River fall chinook could disappear in the years between enabling legislation and the actual construction of the Snake River hatchery. The Snake River Fall Chinook Egg Bank Program resulted from concern that the stock would continue to decline during the hatchery construction period;

in fact, the hatchery site had not even been identified. The egg bank concept entailed rearing and release of Snake River fall chinook in a lower river hatchery where the outmigrant juveniles and returning adults would avoid mortality associated with passage at Columbia and Snake River dams.

The first effort to establish an egg bank of Snake River fall chinook resulted from a National Marine Fisheries Service (NMFS) proposal presented to, and approved by the Columbia Basin Fish and Wildlife Council (Council). The project was funded by the Pacific Northwest Regional Commission for about \$66,700.

Adults were captured at Little Goose Dam by NMFS employees who incubated the eggs at Lower Granite Dam (Figure 1). Eventually, eggs were transferred to Bonneville FH (operated by Oregon Department of Fish and Wildlife; ODFW), and were hatched and reared there. After much deliberation, the fish were coded-wire tagged and sent to Kalama Falls FH (operated by Washington Department of Fisheries; WDF), and released. The results of this first egg bank effort were not rewarding but the concept had been established in practice.

By June 1977, the Council had discussed various problems with the trapping facility at Little Goose Dam and suggested Ice Harbor Dam as a potential trapping site. The Artificial Production Committee (APC), a standing committee of the Council, was assigned to develop an egg bank plan for 1977. At that time the Idaho Cooperative Fish Research Unit (ICFRU) volunteered to conduct the trapping operation, based upon their previous experience trapping adults at hydropower projects. The NMFS was selected to do the adult transport and

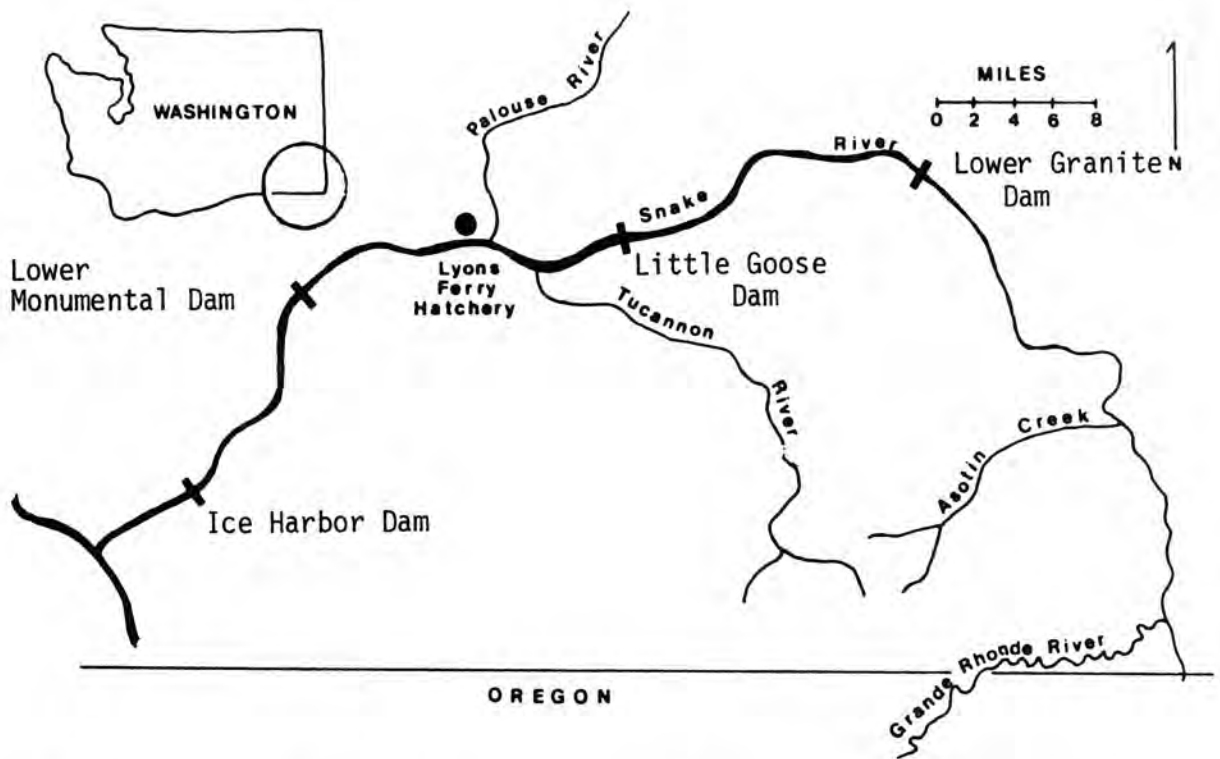


Figure 1. Snake River Basin, showing location of hydroelectric dams, and Lyons Ferry Fish Hatchery.

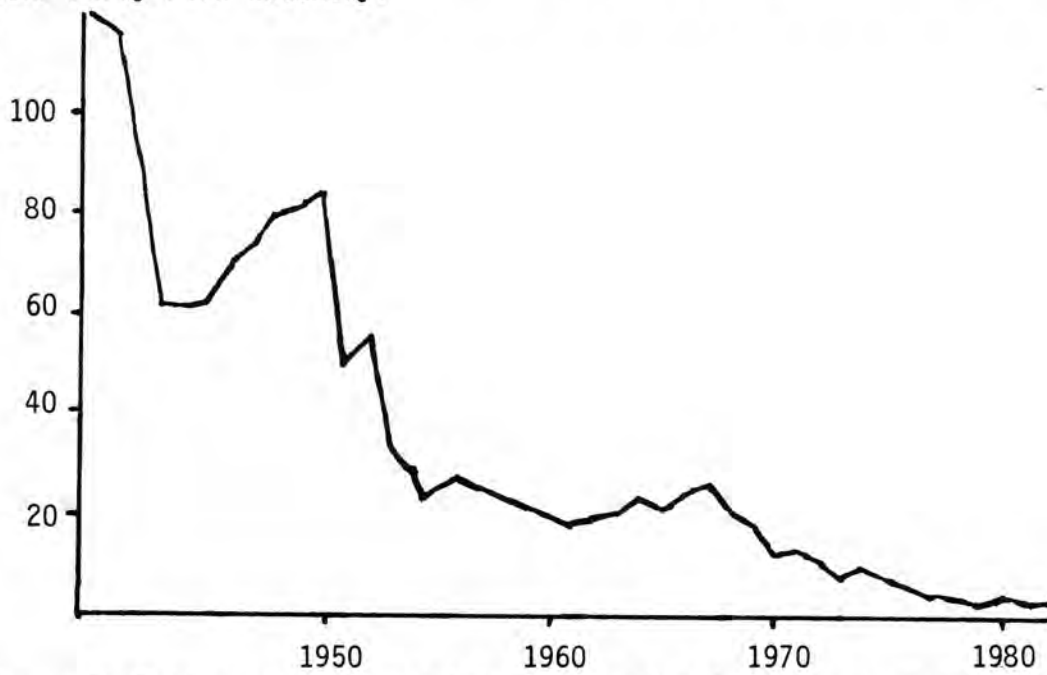


Figure 2. Estimated numbers of fall chinook salmon entering the Snake River during the period 1940 to 1980 (adapted from Fulton 1968, USACE 1964).

provide funding for hatchery rearing. The adult holding facility was first to be Klickitat FH (operated by WDF), but was later changed to Tucannon FH (Washington Department of Wildlife; WDF), because of logistical considerations. Klickitat FH was retained as the juvenile rearing facility, however, because of its adequate rearing area and water supply. Kalama Falls FH was again chosen as the release site. All Snake River stock fall chinook were marked (ventral-fan clip) to allow discernment from the Kalama River stock as adults. This strategy continued in 1978 and 1979 (Table 1). From 1979 to 1986, marked Snake River stock fall chinook returned to Kalama Falls FH, and were used in addition to the fish trapped at Ice Harbor for the Egg Bank Program. Return rates for these release groups, based upon the count of marked fish spawned at Kalama Falls FH, ranged from 0.07 to 0.16 percent. In a 1978 Columbia Basin Fisheries Technical Committee (Tech. Comm.) meeting, the U.S. Fish and

Wildlife Service (USFWS) proposed that Hagerman National Fish Hatchery be included in the Snake River Fall Chinook Egg Bank Program. Plans were to rear 100,000 subyearling smolts for release in the middle Snake River (upstream of Lower Granite Dam). The rationale for this decision was to maintain a second source of this stock in case a major disease, or some other factors, would decimate the primary source. The Hagerman FH plan was approved by the Tech. Comm. and by the APC; transfer of eyed eggs from Tucannon FH began in 1978. At about this time, the Council stipulated that the Egg Bank Program would trap 400 fish or 50% of the run, whichever was lowest. The APC recommended that the egg take would be split evenly between the WDF (Kalama Falls FH) and the USFWS (Hagerman FH) programs. In 1980, Dworshak FH began receiving adults for the USFWS Hagerman FH program (Table 1).

Table 1. Adult holding, juvenile rearing, and smolt release locations for fall chinook salmon under the Snake River Egg Bank Program for the 1977 - 1985 brood years.

| Brood Year | Adult Holding | Juvenile Rearing | Smolt Releases |
|------------|------------------|------------------|--------------------|
| 1976 | Little Goose Dam | Bonneville FH | Kalama River |
| 1977 | Tucannon FH | Klickitat FH | Kalama River |
| 1978 | Tucannon FH | Klickitat FH | Kalama River |
| 1979 | Tucannon FH | Hagerman FH | Middle Snake River |
| | | Klickitat FH | Kalama River |
| 1980 | Tucannon FH | Hagerman FH | Middle Snake River |
| | Dworshak FH | Klickitat FH | Kalama River |
| | | Hagerman FH | Middle Snake River |
| 1981 | Tucannon FH | Klickitat FH | Kalama River |
| | Dworshak FH | Hagerman FH | Middle Snake River |
| 1982 | Tucannon FH | Klickitat FH | Kalama River |
| | Dworshak FH | Hagerman FH | Middle Snake River |
| 1983 | Tucannon FH | Klickitat FH | Lower Snake River |
| | Dworshak FH | Hagerman FH | Middle Snake River |
| 1984 | Lyons Ferry FH | Lyons Ferry FH | Lower Snake River |
| | | Hagerman FH | Middle Snake River |
| 1985 | Lyons Ferry FH | Lyons Ferry FH | Lower Snake River |

Beginning with the 1980 brood year, two factors occurred which improved the future of the Snake River Egg Bank Program: (1) funding for the trapping operation was provided by the USFWS under the Lower Snake River Compensation Plan (LSRCP), and (2) construction of the Snake River fall chinook hatchery at Lyons Ferry was underway. The WDF Lyons Ferry FH was completed in time to handle the 1984 brood year adults, eliminating the necessity to handle adults at either Tucannon or Dworshak hatcheries. The ICFRU continued the trapping operation at Ice Harbor Dam, but was now supplying the adults directly to Lyons Ferry FH (Ringe and Bugert 1990; Table 1).

Lyons Ferry FH, located on the lower Snake River (Figure 1), then began the

process of broodstock building. In 1984, during its first year of operations, Lyons Ferry FH began receiving all eyed eggs from the Snake River stock fall chinook spawned at Kalama Falls FH. This transfer would continue through 1986, the last year significant numbers of Snake River stock fall chinook returned to Kalama Falls FH. The 1982 brood was the last year the Snake River stock fall chinook were released from Kalama Falls FH (Table 2). Eggs supplied from Kalama Falls FH contributed 25%, 62%, and 56% to the Lyons Ferry FH eggtake in 1984, 1985, and 1986, respectively. Beginning in 1986, voluntary returns of the 1983 brood fall chinook released from Lyons Ferry FH added to the broodstock building process.

Table 2. Contribution of Snake River fall chinook adults and jacks to Lyons Ferry FH from 1984 through 1988.

| Year | Collection Point | Number Collected | |
|------|------------------|------------------|-------|
| | | Adults | Jacks |
| 1984 | Lyons Ferry FH | 0 | 0 |
| | Ice Harbor Dam | 663 | 97 |
| | Kalama Falls FH | 220 | 10 |
| 1985 | Lyons Ferry FH | 0 | 4,070 |
| | Ice Harbor Dam | 589 | 90 |
| | Kalama Falls FH | 952 | 0 |
| 1986 | Lyons Ferry FH | 245 | 1,125 |
| | Ice Harbor Dam | 212 | 23 |
| | Kalama Falls FH | 576 | 0 |
| 1987 | Lyons Ferry FH | 1,654 | 543 |
| | Ice Harbor Dam | 1,613 | 47 |
| 1988 | Lyons Ferry FH | 327 | 1,053 |
| | Ice Harbor Dam | 1,076 | 6 |

Most of the 1979 through 1984 broods of Snake River stock fall chinook reared at Hagerman FH were planted at various locations in the middle Snake River between Hells Canyon Dam and Lower Granite Dam. Several groups were also transported for release below Bonneville

Dam. A portion of each brood year, except the 1982 brood, was coded-wire tagged (Table 3). Adult survival and contribution rates ranged from 0.01 to 0.47 percent; return rates to the Snake River ranged from 0.01 to 0.24 percent.

CURRENT STATUS

Trapping of fall chinook at Ice Harbor Dam continues, but is now conducted by WDF to supplement voluntary returns to the Lyons Ferry FH rack. All releases of Snake River stock fall chinook since 1985 have been from Lyons Ferry FH (Bugert et al. 1989).

Under contract of the USFWS, WDF geneticists collected and compared electrophoretic samples from the 1986 Snake River stock fall chinook that returned to Kalama Falls FH and those that returned to the Snake River. They found no evidence of genetic difference between these two groups, based upon

examination of allele frequencies of 30 variable loci (Seidel et al. 1988).

We feel the Snake River Fall Chinook Egg Bank Program was successful because of two factors: (1) smolts released from Kalama Falls FH and into the middle Snake River from Hagerman FH contributed substantially to the broodstock building process during the first three years Lyons Ferry FH became operational, and (2) genetic integrity of a stock once considered for inclusion on the federal endangered species list was maintained as a result of cooperative ventures between several agencies, and careful broodstock management by several hatcheries.

Table 3. Location, size, and number of Snake River stock fall chinook released by Hagerman National Fish Hatchery under the Snake River Fall Chinook Egg Bank Program. Data are presented by coded-wire tag mark and recovery rates.

| Brood Year | Tag Code | Number Tagged | Release Location | Size at Release (fpp) | Number Recovered | Percent Recovered | Total Release | Expanded Recovery |
|------------|----------|---------------|-----------------------------------|-----------------------|------------------|-------------------|---------------|-------------------|
| 1978 | 05-04-20 | 52,000 | Below Bonneville Dam | 84 | 56 | 0.107 | 93,000 | 99 |
| | 05-04-21 | 45,361 | Near Asotin Cr. | 92 | 6 | 0.013 | 45,361 | 6 |
| 1979 | 05-05-27 | 58,100 | Near Asotin Cr. | 57 | 174 | 0.299 | 165,500 | 496 |
| | 05-05-28 | 56,000 | Below Bonneville Dam | 59 | 24 | 0.043 | 56,000 | 24 |
| 1980 | 10-22-10 | 55,400 | Above L. Granite Dam | 34 | 174 | 0.314 | 120,157 | 377 |
| | 10-22-11 | 55,700 | Below Bonneville Dam | 51 | 156 | 0.280 | 61,134 | 171 |
| 1981 | 05-10-22 | 78,300 | Near Asotin Cr. | 37 | 350 | 0.447 | 394,395 | 1,763 |
| | 05-10-23 | 80,421 | Above L. Granite Dam | 37 | 375 | 0.466 | 80,721 | 376 |
| 1982 | None | ----- | Near Grande Ronde River | 44 | --- | ----- | 78,900 | |
| 1983 | 05-13-54 | 59,300 | Near Grande Ronde River | 53 | 113 | 0.191 | 427,191 | 814 |
| 1984 | 05-13-53 | 54,925 | Near Asotin Cr. & Grande Ronde R. | 44 | 96 ¹ | 0.175 | 128,229 | 224 |

¹/Age 5 recoveries not included

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**ENNIS NATIONAL FISH HATCHERY-
GENETICS/BROODSTOCK**

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What causes the loss of genetic variation?

GENETIC DRIFT: the random genetic changes which occur from one generation to the next if too few parents contribute gametes to the next generation. The result is that the genetic make-up of each successive generation is less variable because alleles are lost. An extreme example of drift is when a very small number of parents are used in any given generation. This produces a bottleneck. The effects of using a small number of parents to start a population are called "founder effects."

INBREEDING: the mating of related individuals. The more closely related these individuals are, the less variable they are. In small populations, inbreeding may become a problem. The odds of mating relatives are high.

INTENTIONAL SELECTION: genetic selection reduces the genetic variation at all genes, not just those affecting the targeted trait. If the selected population is small, you may select for individuals that are closely related, thus increasing the rate of drift and inbreeding.

UNINTENTIONAL SELECTION: domestication.

It is apparent that loss of variation in our fish stocks is undesirable. It will also become apparent that preventing the loss of genetic variation will mean a change in the way we do business. Some of the changes may have an immediate beneficial effect, but the true benefits of maintaining genetic variation will become apparent in the future.

How can the loss of genetic variation be minimized?

COLLECT BASELINE DATA:

-In order to determine the genetic health of a brood stock, it is necessary to collect baseline information. This can be done by contracting for an electrophoretic analysis of each species and strain, asymmetry counts of each species and strain, or any other of the methods that have become widely accepted (ex. DNA analysis).

PLAN YOUR PROGRAM:

-Avoid intentional selection. Once again, remember that you can't select for performance in the wild.

-Eliminate unintentional selection as much as possible. Do any number of innovative things to keep those "wild" fish alive on the hatchery until they can contribute to the next generation.

-When testing indicates a need for it, go back to the founding population for an infusion of genes. Allendorf and Leary have suggested that a 10% infusion of genes from the founding population every 10 years (3 generations) may be sufficient to minimize domestication. This is a rule of thumb to be supported by electrophoretic testing.

-There is no substitute for long range planning. If you plan your moves step by step to avoid selection, inbreeding, and drift, you can avoid major genetic problems in the future.

REDUCE LOSS OF VARIABILITY:

-Avoid inbreeding. The closer the relationship, the less variable offspring will be. Using males and females of different ages will avoid brother X sister matings, and prevent reproductive isolation of year classes

-Mating ratio. Mate one male with one female. Mated pairs enhance the effective population size. One half of the genetic material comes from the male and one half from the female. If 1:1 ratios are not possible, try at least 60:40 ratios.

-Equal contribution. Use an equal number of eggs from each paired mating. One female may have 5,000 eggs and another only 1,500. It is important that each mated pair contributes an equal number of offspring to the next generation. The contribution of eggs from each take should be in proportion to the total number of ripe females on that date.

-Number of spawning takes. Spawning time is highly heritable. Fish that spawn on the same day may be related; therefore, future broodstock should be represented by eggs from at least 60% of the total egg takes. Also, the beginning and tail end of the spawning season represents less genetic variation than the peak spawning time so use caution when deciding which takes to use for future brood.

-Outcrossing. A 10% infusion of genes from the founding population every 10 years or so (3 generations) is recommended by Allendorf and Leary; however, this needs to be supported by electrophoretic information. "Strains" can be crossed to provide an infusion of variability, but get help from professionals. Make sure what crosses are necessary in advance of doing the actual work. Protect parental groups from genetic contamination.

-Selection. Remember unintentional selection is taking place all the time in your hatchery. Guard against it and reduce it by keeping all the fish alive except those that are obviously deformed.

-Mating schemes. Mate individuals at random. Only obviously deformed fish should be excluded from contributing to future generations. Avoid selecting for early maturity by mating individuals at an age when 90% of the population becomes sexually mature. If there is any doubt about sperm quality of one male, use another male to fertilize the eggs from that female.

EFFECTIVE BREEDING NUMBER:

The effects of unequal numbers of males and females on reducing effective population size below actual size can be estimated using this formula:

$$NE = \frac{4 (NF) * (NM)}{(NF) + (NM)}$$

Where: NE= Effective Number
NF= Number of Females
NM= Number of Males

Consider founding a hatchery stock using 99 females and 1 male. Remember that each sex contributes one half of the genetic material to the next generation.

$$NE = \frac{4 (99) * (1)}{(99) + (1)} = 3.96$$

The inbreeding and genetic drift for a population of 99 females and 1 male is greater than that produced by a population composed of 2 males and 2 females!

RATE OF LOSS:

The rate of genetic loss per generation by drift can be defined as:

$$\frac{1}{2 (NE)}$$

Examples:

NE = 4 The rate of genetic loss in one generation is 12.5%

NE = 50 The rate of loss is 1%

NE = 250 The rate of loss is 0.2%

Effective population size is inversely related to the rate of loss by genetic drift.

GENETIC MAINTENANCE OF A HATCHERY STRAIN OF TROUT:

The recommended number of paired matings to maintain a broodstock is 250. The minimum number for maintenance is 100 pairs. These numbers are referring to the number of fish pairs which contribute gametes to a future generation. In order to have 250 contributing pairs of fish, a hatchery must allow for: 1) the average annual mortality; 2) the mortality from fish health sampling; and 3) the percent of fish that are not expected to contribute because

they are either immature, barren, or physically deformed. For most hatcheries, this translates to a broodlot containing 450 to 500 pairs of fish to ensure genetic contribution for 250 pairs.

Taking an equal number of eggs from each mated pair for future broodstock will help each pair contribute equally to the next generation. (Remember, the males can only be used once too!)

Since the time of spawning is highly heritable, and the genetic variation between spawning dates is significant, it is strongly recommended that a minimum of 60% of the spawning dates be represented in those eggs taken for a future generation. If 10 spawning dates are expected, 6 of those should be used to select future broodstock. Using all 10 takes would be the most desirable.

The percent of progeny from each take contributing to future broodstock is relative to the total number of females ripe on those dates that matings for future brood were made.

For Example:

| Spawn Dates | May 1 | May 7 | May 14 | May 21 | May 28 |
|--------------|-------|-------|--------|--------|--------|
| Females Ripe | 75 | 200 | 500 | 300 | 100 |
| Females Used | 0 | 100 | 100 | 100 | 0 |

Total number of females ripe on the days brood eggs were taken = 1,000
May 7 percent of total = 20%
May 14 percent of total = 50%
May 21 percent of total = 30%
(Disregard May 1 and May 28 since no brood were taken then.)

If 5,000 eggs are needed for future brood, 20% would come for the paired matings on May 7 (1,000), 50% would come from May 14 (2,500), and 30% would come from May 21 (1,500). This scheme will help insure genetic variation while maintaining the time of spawning.

BROODSTOCK

The protocol for bounding a broodstock is vital to the genetic survival of that stock.

Improper methods of founding future generations will result in continued loss of variation until eventually there will not be enough variation to allow adaptation to changing environmental conditions.

You can expect that "proper methods" for founding fish stocks will always be less convenient than the "easy way", but please consider the end result before making that final decision to do it the easy way!

FOUNDING BROODSTOCK FROM A FREE ROAMING POPULATION:

It is desirable to found a new population from a minimum of 100 pairs of fish. More would be better, although using more than 250 pairs is probably not practical nor necessary. Since you may have trouble collecting enough fish in any one season, it

should be done over a period of 3 successive years. Be sure to develop a scheme to cross all 3 year classes with each other as they mature. As a reminder, paired matings means just that: 1 good male X 1 female. (If milt is watery or coagulated, use another male to be sure!)

If possible, collect gametes over the course of several days or weeks because females spawning very close together have a greater likelihood of being related. Spreading out collection dates will enhance the gene pool of the founding generation.

Use an equal number of eggs from each mated pair to avoid the genetic drift caused by unequal contribution of offspring from each mating.

Regardless of how you manage a wild stock at the hatchery, unintentional selection adapts it to the hatchery environment. This will result in improved performance in the hatchery, but will likely cause reduced performance in the wild. Therefore, the hatchery population should be crossed with the wild population every few generations when electrophoresis and other diagnostic methods indicate it is necessary. A monitoring program should check the population every 10 years or so. In order to tell if you stock has changed genetically, the first thing you must do is get baseline information from the original stock!

FOUNDING A BROODSTOCK FROM DOMESTIC (CAPTIVE) SOURCES:

Most domestic hatchery broodstocks have been in captivity for a long time and have probably undergone substantial genetic change. It is important to prevent further erosion of the genetic variation presently available in these stocks.

Do baseline electrophoresis and asymmetry counts on the stock being used as founders.

Use paired matings from at least 60% of the available spawning dates. (100% is ideal of course).

Use a minimum of 250 paired matings to ensure good genetic representation.

Select an equal number of eggs from each paired mating.

Provide eggs to the founding hatchery for 2 or 3 generations and make sure those year classes are crossed.

Periodically return to the original population for an infusion of genetic material. This necessity can be determined by electrophoretic testing and other accepted methods of genetic monitoring that may be developed.

APACHE TROUT CULTURE PROGRAM

By

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The Apache trout (*Oncorhynchus apache* formerly *Salmo apache*), a native trout of Arizona, is limited to a small number of high mountain streams, in the eastern central portion of the state. Its existence in the White Mountains of Arizona has been known since 1873, (at which time, it was thought to be a cutthroat trout), but has been scientifically described as a distinct species only as recently as 1972.

Historically, the range of the species included the upper Salt River (Black River and White River), the San Francisco River (Blue River), and the Little Colorado River. In the early 1950's, however, the White Mountain Apache Indian Tribe noted a substantial reduction in numbers of this fish on the Fort Apache Indian Reservation. Consequently, in 1955, the tribe restricted and/or eliminated much of the angling pressure on the Apache trout on the Reservation. Since that time, virtually all other populations have been extirpated. The Apache trout was listed as 'threatened with extinction' under the Endangered Species Conservation Act of 1969, and then 'endangered' with the passage of the Endangered Species Act of 1973. In July of 1975, although approximately only 30 isolated populations of Apache trout remained, the status of the species was downlisted to 'threatened'. Currently, less than 15 self-sustaining populations are known.

Apache trout populations have declined for a number of reasons, including:

- 1) the loss of suitable habitat
- 2) overfishing
- 3) competition from exotics (brooks, browns rainbows, and cutthroats have been stocked in Apache trout waters for over 50 years)

- 4) hybridization with exotics (primarily rainbow)

APACHE TROUT CULTURE HISTORY

- | | |
|-----------|---|
| 1963 | Arizona Department of Game and Fish collected 82 fish from Ord Creek. Spawning efforts resulted in 99 fry which became future broodstock. |
| 1966-1974 | Very little success. Hard to get fry to feed. |
| 1974 | All fish stolen. |
| 1975 | 118 fish collected from Soldier Creek. Progeny held for future broodstock. |
| 1978-1980 | Little success. Egg mortality 70% |
| 1981 | Program abandoned. |

The U.S. Fish and Wildlife Service picked up the Apache trout program in 1983. In 1983 and again in 1984, personnel from the Williams Creek NFH collected eggs from wild Apache trout on the Fort Apache Indian Reservation. This effort resulted in approximately 800 fry which were held for future broodstock.

Apache Trout Production At Williams Creek NFH

- | | |
|------|---|
| 1986 | Apache trout from the 1983 year class were successfully spawned on station. 60,000 eggs were taken which resulted in 1,400 fry. |
| 1987 | Two year classes of broodstock were spawned. 22,150 fingerlings were produced. |

1988 90,000 Apache trout fingerlings produced.

1989 175,000 Apache trout produced.

Innovations in nutrition, spawning techniques, and general fish culture have allowed for a successful Apache trout culture program At Williams Creek NFH. The station now supports four year classes of broodstock and will have released approximately 300,000 Apache trout over the past three years. Future production goals call for the release of almost 500,000 per year.

The Apache trout culture program at the Williams Creek NFH is an important part of the Apache trout recovery plan which was drafted in 1983 by the Apache trout recovery team. The recovery plan is a joint effort involving the U.S. Fish and Wildlife Service and the White Mountain Apache Tribe, the Bureau of Indian Affairs, the Arizona Department of Game and Fish, and the U.S. Forest Service. The primary objective of the recovery plan is to remove the Apache trout from the Threatened Species List. However, the ultimate goal is to return the Apache trout to its role as the dominant game fish in waters throughout its historical range.

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JOCKO RIVER TROUT HATCHERY

Sibley A. Malee
Montana Department of Fish, Wildlife and Parks

Montana has it all, whether its winter, summer, spring or fall. We have many sports related activities, but none compare to fishing.

The state owns and operates nine hatcheries which produce 10.2 million trout each year to release across the state as fingerlings, catchables and retired broodstock. Four of the nine hatcheries are brood facilities. Salmonid broodstocks managed are: the McBride cutthroat, Westslope cutthroat, and the Arlee rainbow.

Montana's trout streams are the bread and butter of the states fisheries' resources. Our 20,000 miles of trout streams support approximately 1.25 million angler days of use per year. Trout streams, including 541 miles of Class I Blue Ribbon streams, are managed almost exclusively for wild trout. The hatchery production is directed to lakes and reservoirs.

Wild trout management is built on habitat preservation. Streams that have adequate flows, good water quality, and stable beds and banks will produce adequate fish populations naturally. Special regulations with more restrictive limits are sometimes used to maintain good fishing.

There are approximately 1900 cold water lakes providing one million angler days of recreation per year. Anglers seek trout and salmon in alpine lakes, large reservoirs and western Montana lowland lakes. Although naturally reproducing populations are found in some lakes, the majority of lakes require periodic stocking to maintain fishable populations. Rainbow trout is the species most commonly stocked. Six strains of rainbow trout are being tested to take advantage of specific traits. But most lakes and reservoirs have little or no potential for natural reproduction and are stocked with our domestic Arlee rainbow that performs well in put-grow-take situations. Kokanee salmon, both wild and stocked provides significant cold water fishing opportunities in northwest and west central Montana.

Current and past brood management has been the result of monitoring performance of feed conversion, %eye up, fecundity, fry survival, and observed changes in the spawning range and peak. Until 1984, the future brood eggs were kept from one spawning take as the hatchery's select eggs. These fish were to represent the total population when mature. Hatchery personnel observed the range and peak of the spawning season becoming increasing more confined from 1978 to 1984, an indication of inbreeding. (Graphs 1,2) In 1984, brood recruitment techniques were radically changed. Eggs were kept from all but one of the spawning weeks and were proportioned to form a more normal bell curve as in 1978.(Graph 3) Inherit in keeping too many eggs from the tailends of a spawning period is the potential that the total heterozygosity of the population can be reduced. In the winter of 1985, the percent heterozygosity of the different weekly takes was measured by Dr. Robb Leary, University of Montana, Populations Genetics Lab. They concluded:

1. some significant differences in individual loci from one egg take to the next that weren't related to time of spawning.
2. loci normally polymorphic were found to be so except in the last egg take.
3. average heterozygosity was significantly lower in the first and last eggs takes.

The early and late takes are significantly different than those in the middle of the spawning season. This suggests that progeny produced in the middle of the spawning season has more genetic variation than progeny produced either early or late in the spawning season. Furthermore it suggests that spawning time is influenced by many genes spread throughout the genome of rainbow trout.

By keeping future brood eggs from 5-7 takes, our spawning curve in 1987 is now almost identical to that in 1978 (Graph 4). Our current breeding scheme is primarily addressing two factors that can cause loss of genetic variation;

1. the number of brood fish contributing genes to future broodstock.
2. selection for spawning time.

But how do you know how many individuals

from each spawning date should be selected for inclusion in the future brood stock? And the numbers of future brood eggs to be kept? The number of fish kept from each date should be proportional to the number of females spawned that day, relative to the total number spawned from which future brood stock is to be chosen. For example: Total number of fish to be spawned is 1000. Your first take you spawned 100, the second 200, the third 400, the fourth 200, and the last 100. Disregard your first and last takes.

| Spawn date | 9-15 | 10-1 | 10-15 | 11-1 | 11-15 | 12-1 | 12-15 |
|-------------------------------------|------|------|-------|------|-------|------|-------|
| Females ripe | 75 | 100 | 200 | 400 | 200 | 100 | 50 |
| Females kept for future brood fish* | 0 | 50 | 100 | 200 | 100 | 50 | 0 |

* we keep 500; minimum should be 250 that will spawn the following year.

Total number of females to be kept for future brood fish:
 09-15 = no future fish kept
 10-01 = 100 females or 10% of 1000 = 50 future brood fish
 10-15 = 200 females or 20% of 1000 = 100 future brood fish
 11-01 = 400 females or 40% of 1000 = 200 future brood fish
 11-15 = 200 females or 20% of 1000 = 100 future brood fish
 12-01 = 100 females or 10% of 1000 = 50 future brood fish
 12-15 = no future fish kept 500 future brood fish

For selecting brood eggs

| Spawn date | 9-15 | 10-1 | 10-15 | 11-1 | 11-15 | 12-1 | 12-15 |
|-------------------------------------|------|------|-------|------|-------|------|-------|
| Females ripe | 25 | 50 | 100 | 200 | 100 | 50 | 25 |
| Females Used for future brood eggs* | 0 | 50 | 100 | 200 | 100 | 50 | 0 |

* total number should be minimum of 250;(we use 500 pairs)

Total number of used females ripe = 500
 09-15 = no future brood eggs taken
 10-01 = 50 females or 10% = 1000 brood eggs taken
 10-15 = 100 females or 20% = 2000 brood eggs taken
 11-01 = 200 females or 35% = 3500 brood eggs taken
 11-15 = 100 females or 25% = 2500 brood eggs taken
 12-01 = 50 females or 10% = 1000 brood eggs taken
 12-15 = no future brood eggs taken
 10,000 total brood eggs taken

This scheme will help to insure genetic variation for your broodstocks.

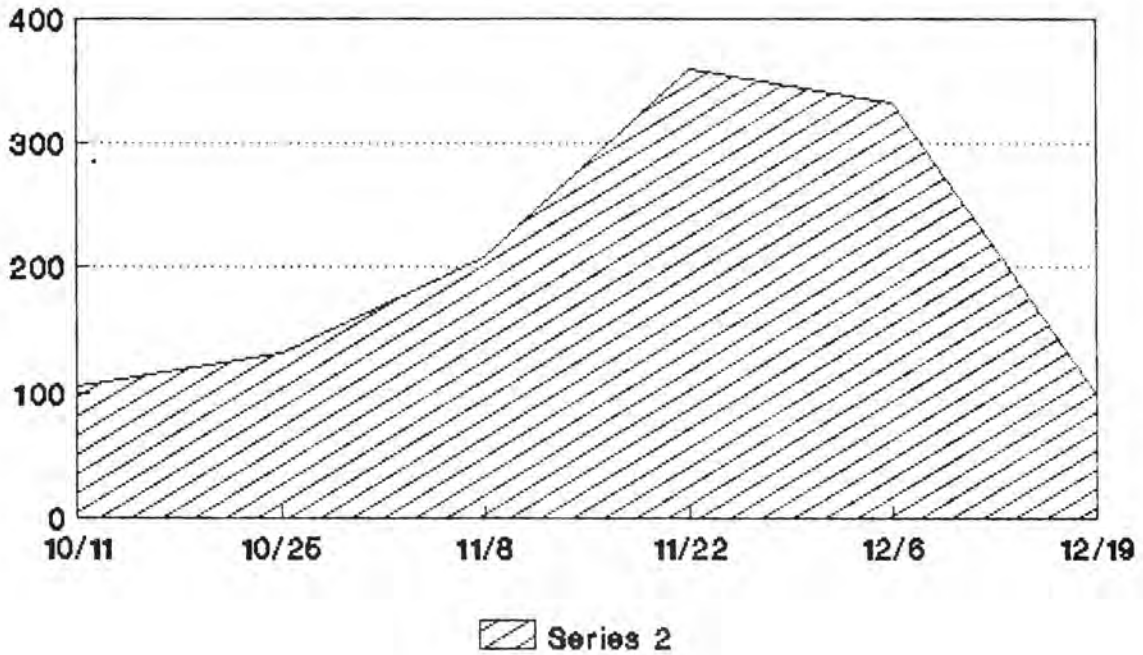
We cross 4 year old females with 3 year old males for future brood eggs. Sperm is collected into separated vials, each male contributing equal amounts. After fertilization, eggs are water-harden in a 75 ppm betadine solution for 30 minutes, then put away into Montana hatching boxes.

They are treated daily with a 1250 ppm formalin treatment for 15 minutes to prevent fungus.

Eggs are enumerated using the Von Bayer method. Eggs are packed in wood-framed trays and shipped with in Montana and to other state and federal hatcheries. The Jocko River Trout Hatchery produces 8 million Arlee rainbow annually from Aug 15 to Jan 1.

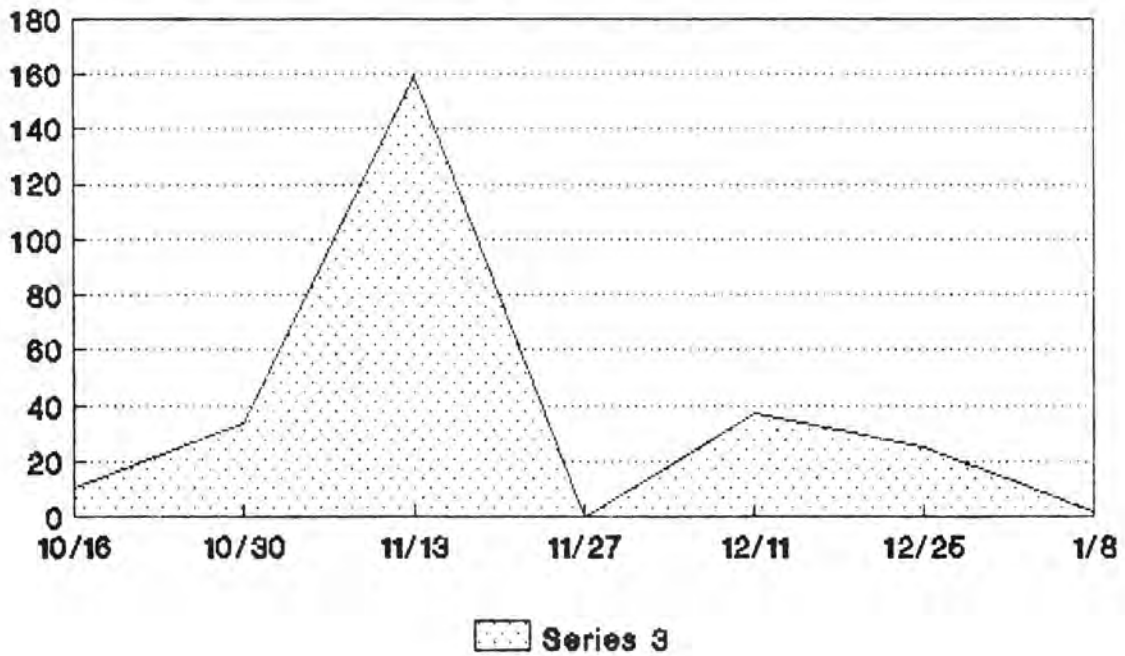
Graph 1

Arlee Rainbow Spawning Curve - 1978



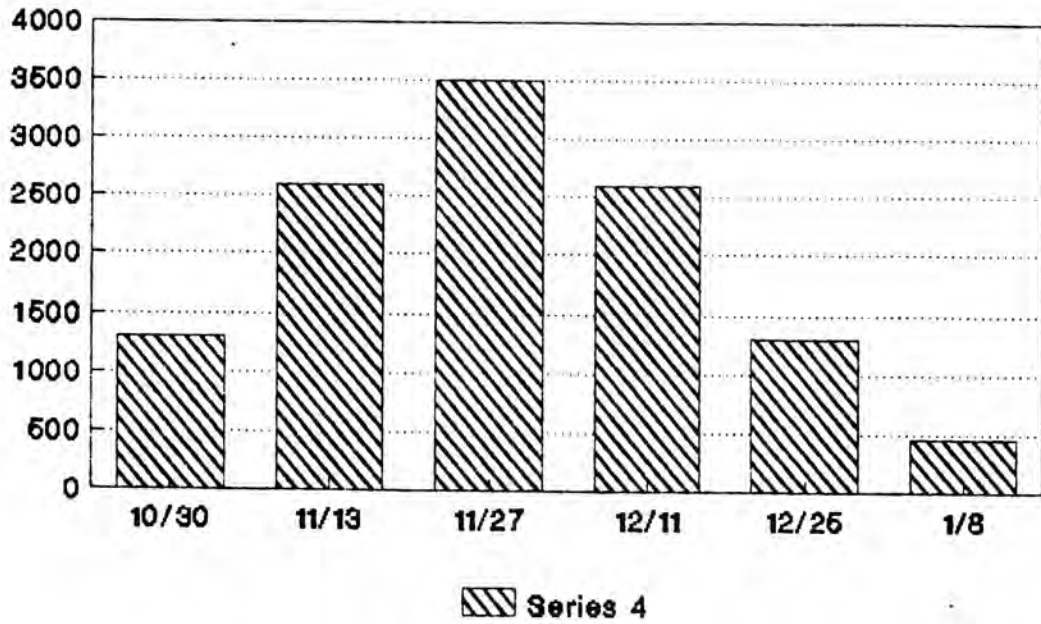
Graph 2

Arlee Rainbow Spawning Curve - 1984



Graph 3

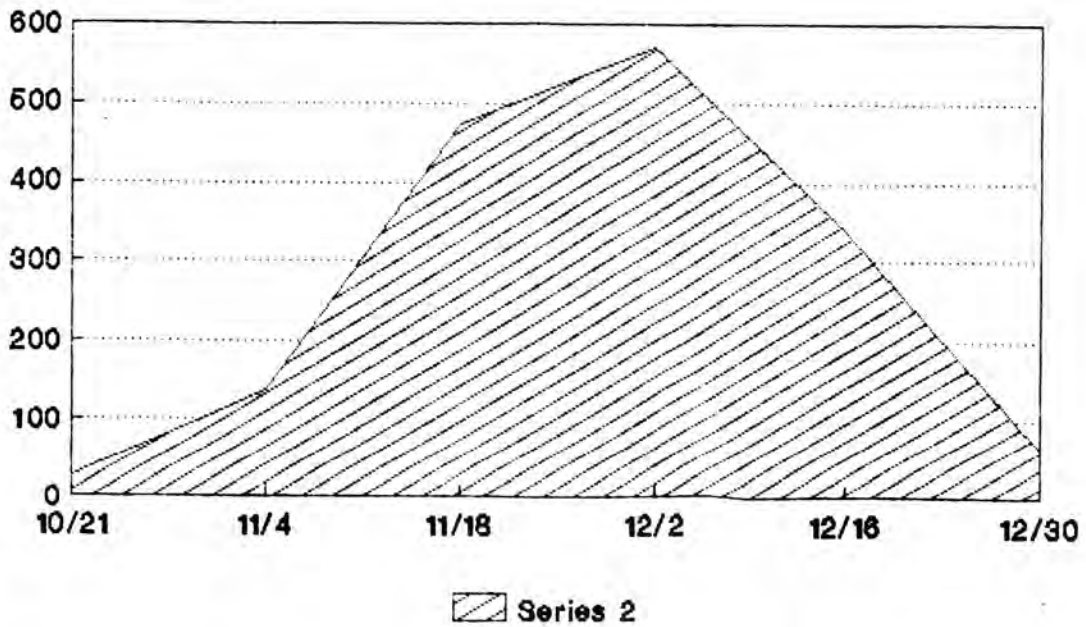
Arlee Rainbow Brood Recruitment



1984

Graph 4

Arlee Rainbow Spawning Curve -1987



INJECTING PURE OXYGEN INTO SEALED COLUMNS- PERFORMANCE

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ABSTRACT

Sealed columns have considerable potential in aquaculture. They are simple, effective devices which can economically add oxygen to water. Increasing oxygen content of water enables fish culturists to increase their fish loads and improve the rearing environment. A series of tests with water flows of 770, 481, 385, and 288 gpm versus gas/liquid (G/L) ratios of 0.5, 1.0, 2.0, 3.0, and 4.0% was conducted at each of four hatcheries. Actual water flow and G/L ratios varied slightly from the nominal values. At one hatchery, Delta DO (amount of oxygen added to the water) ranged from 6 to 26 mg/L as G/L ratios increased from .69 to 5.3. Results at the other three locations followed the same trends. Tests also indicated that there were no differences in performance of a column with 3 feet versus 5 feet of packing over the range of flows tested. Two types of packing were compared but little difference was noted. Operation of column with packing was compared to that of one using a spray nozzle. While results were quite similar, the nozzle performed best at the higher water flows tested.

INTRODUCTION

The demand for hatchery fish often exceeds the number that can be safely supplied by many federal and state fish hatcheries. One solution is to increase the carrying capacity and improve the environment in which the fish are raised rather than to construct more facilities. Recent advances in technology make the injection of oxygen a viable alternative to the construction of additional rearing

units (Speece 1981). This can also improve the environment by reducing nitrogen saturation (Westers, personal communications).

Sealed columns were first used at hatcheries in Michigan to reduce nitrogen saturation and increase oxygen content of the water (Westers, personal communication). In a previous study (Dwyer et. al. 1988), it was shown that the use of sealed columns is an efficient means of injecting oxygen into water. Much of the data collected in that study were from a 4 inch column in the laboratory at lower water and oxygen flow rates than practical for fish culture.

This project was completed using 24 inch diameter production size columns to determine operational characteristics as well as to evaluate the effects of column height, media, and to compare operation with media or a nozzle.

Watten et. al. (1988) developed a model to define the affects of varying parameters such as nitrogen and oxygen on the column oxygen adsorption efficiency. This work shows that based on the model, oxygen absorption rate can be predicted. Our studies compared data collected with those predicted by the model.

OBJECTIVES

- 1). Determine oxygen absorption efficiency rates using a 24 inch diameter sealed column at a series of oxygen and water flows to establish practical flow rates. This was done at four different locations

in order to account for differences in water quality and to assure that trends remained constant.

- 2). Determine the effect of column length (7 ft and 5 ft columns with 5 ft versus 3 ft of packing) as described above in objective 1.
- 3). Compare spray nozzles and media to determine differences in oxygen absorption efficiency.
- 4). Compare actual data obtained from our test with predicted values obtained from the model described by Watten et. al. (1988).

This information will be of value to design engineers and hatchery managers when building sealed columns to meet specific needs. Needs may include nitrogen removal, improved oxygen absorption, or both.

METHODS

Water and oxygen flow rates shown in Table 1 were used in all tests with both columns. Water flow rates tested were 770 gpm (1 L/m per square centimeter of cross sectional column area as recommended by Owsley (1981) for packed columns), 481, 385, and 299 gpm. Several oxygen flow rates were tested at each water flow rate. These were based on G/L ratios expressed as a percent. G/L ratios were calculated as follows: $\{\text{standard gas flow (Lpm)} / \text{water flow (LPM)}\} * 100$ (Watten et. al. 1988). This is the oxygen flow expressed as a percent of the water flow. G/L ratios tested at each water flow were 0.5, 1.0, 2.0, 3.0, and 4.0%

General test using the 24 inch diameter column with 5 feet of packing were conducted in Idaho at the Dworshak, Kooskia, and Hagerman National Fish Hatcheries (NFH) and at the Giant Springs State Fish Hatchery in Montana.

Special tests to evaluate column height and compare packing were conducted at Dworshak NFH. These consisted of a series of measurements at the water and oxygen flow rates shown in Table 1 using the tall and short 24 inch diameter

columns. Both columns were tested using both types of packing.

The spray nozzle was evaluated at Giant Springs State Fish Hatchery near Great Falls, MT. The nozzle used was an industrial product model number 6 R I 65 400 purchased from Spraying Systems Company in Wheaton, IL. It has a 6 inch inlet, the spray angle is 65 degrees, and the capacity is 400 gallons per minute at 3 psi. It is 11.8 inches long by 7.6 inches in diameter and weighs 38 lbs. A series of 14 tests were conducted with the rings and repeated with the nozzle replacing the rings in the columns to compare performance.

Parameters monitored consisted of DO, temperature, delta P, and flow rates for water and oxygen.

RESULTS AND DISCUSSION

General Tests:

Results of the general tests with the 24 inch diameter column containing 5 feet of packing conducted at the Dworshak NFH showed that this column can be used to add oxygen to water very effectively. The incoming water at Dworshak had a DO level of about 10.5 mg/L; after going through the column, the oxygen added to the water (Delta DO) ranged from about 6 to 28 mg/L over the range of water and oxygen flows tested. The Delta DO is plotted against the G/L ratio for each of the water flows tested in Figure 1. This indicates that there is not a great deal of difference in Delta DO at each G/L ratio among the water flow rates. As the G/L ratio increased from 0.5 to 4.0%, the Delta DO showed about a four fold increase.

Oxygen absorption efficiencies were also calculated and were highest at the lowest G/L ratios with a steady decline as the ratio increased from 0.5 to 4.0%. Average absorption efficiencies were 84, 70, 55, 48, and 41% as oxygen flows were increased at each of the G/L ratios (Figure 2). As the absorption efficiency decreased, the Delta DO increased. The data followed the same trends at all locations.

At Kooskia, the mean Delta DO increased from 8.1 mg/L to 22.5 mg/L as the G/L ratio increased from 0.62 to 4.78 and the mean absorption efficiency decreased from 92 to 35% over the same G/L range.

At Hagerman NFH, the mean Delta DO increased from 8.0 to 21.4 mg/L as the G/L Ratio increased from 0.67 to 5.3 and the mean absorption efficiency decreased from 91 to 30%.

TABLE 1

| Water Flow | | Oxygen Flow (L/min) | | | | |
|------------|-------|---------------------|-------|-------|-------|--------|
| gpm | L/min | .05 | 1.0 | 2.0 | 3.0 | 4.0 |
| 770 | 2,914 | 14.57 | 29.28 | 58.28 | 87.40 | 116.56 |
| 481 | 1,820 | 9.10 | 18.20 | 36.40 | 54.60 | 72.80 |
| 385 | 1,457 | 7.28 | 14.57 | 29.14 | 43.70 | 58.28 |
| 288 | 1,090 | 5.45 | 10.90 | 21.80 | 32.70 | 43.60 |

Special Tests:

Column Height-

Under the conditions of this study, there was no obvious difference in performance between the column with 3 feet of packing and the column with 5 feet of packing. These tests conducted at the Dworshak NFH showed that while column height does have an effect, it was not shown between the two column heights tested. Delta DO levels ranged from 7.0 to 25.7 mg/L in the column with 5 feet of packing and from 7.0 to 24.1 mg/L in the short column with 3 feet of Koch ring packing (Figures 3 and 4).

Since there was no effect of column height on Delta DO, there was no effect on oxygen absorption efficiency either. Absorption efficiency ranged from 84.5% at the lowest G/L ratio to 41% at the highest in the 5 feet column, similarly they ranged from 83.8% to 36.3% in the 3 feet column.

Media Comparison-

Two types of media were compared in both columns: these were the 1.5 inch

diameter Koch rings and the 4 X 1.5 inch rings made by Telpack of Boston MA. The two rings are of very different design and we thought that there would be a difference in performance. However, the data showed little overall difference in the two media types tested. The Delta DO ranged from 7.0 to 25.7 mg/L in the 5 feet Koch ring column versus 7.0 to 26.2 mg/L in the 5 feet Telpack column (Figure 5). Results were similar with the 3 feet column. Delta DO ranged from 7.0 to 24.1 mg/L with Koch rings compared to 9.4 to 26.0 mg/L using the Telpack rings (Figure 6).

Nozzle Versus Packing-

The Delta DO ranged from 6.3 to 26.2 mg/L when the column was operated with Koch rings and 7.9 to 22.3 mg/L when the nozzle was installed over the range of G/L ratios tested (Figure 7). Absorption efficiency ranged from 80 to 37% for the column with Koch rings and from 79 to 33% for the column with the nozzle (Figure 8). At the lower water flows, the media seemed to perform slightly better than the nozzle, however, as the water flow increased, the reverse was true.

SUMMARY

- 1). Sealed columns work well for efficiently increasing the DO content of water.
- 2). Columns with as little as 3 feet of packing are quite effective.
- 3). Nozzles are as effective as media at water flow rates near 1 L/min per square centimeter of cross sectional area or 770 gpm with a 24 inch column.
- 4). At a given water flow:
 - A. oxygen absorption efficiency decreases as the oxygen flow rate into the column increases over the range of G/L ratios from 0.05 to 4.0%.
 - B. Delta DO increases as oxygen flow rate into a column increases over the range of G/L ratios of 0.5 to 4.0%.

ACKNOWLEDGEMENTS

This project (877119:Augmented Fish Health Monitoring) was made possible by funding from the Bonneville Power Administration which is greatly appreciated. The columns were constructed and donated by Engineered Products of Corvallis, OR; T.R. Gregg of that company has been very helpful and his assistance and suggestions are also appreciated. Dave Clifford and the staff

at Dworshak NFH assembled the system that was used in all of these tests. We also acknowledge the assistance and patience for the staff and management at Kooskia and Hagerman NFH and Giant Springs State Fish Hatchery.

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DWORSHAK 5 Foot Koch Rings

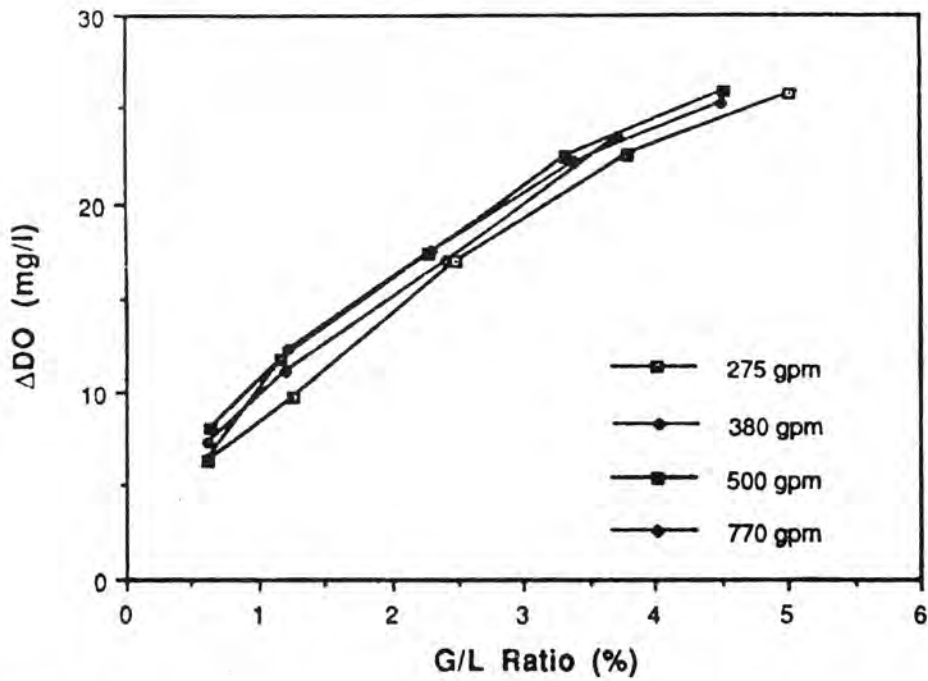


Figure 1. Delta DO or oxygen added (mg/L) to four different water flows through 24-inch sealed column at gas to liquid ratios ranging from 0.63 to 4.7%.

DWORSHAK 5 Foot Koch Rings

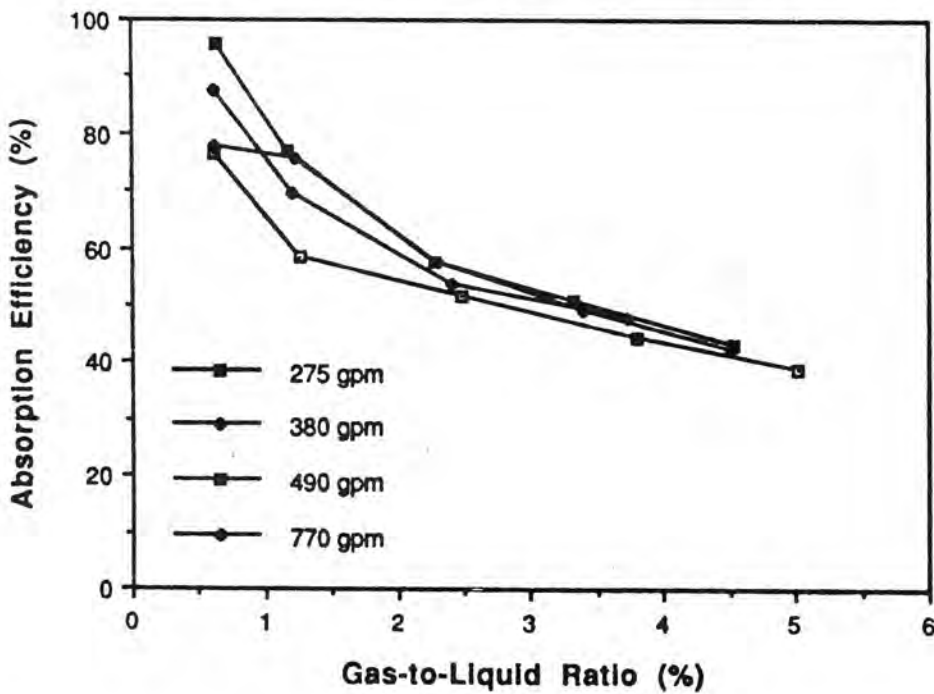


Figure 2. Absorption efficiency (%) at four different water flows through a 24-inch sealed column at gas to liquid ratios ranging from 0.63 to 4.7%.

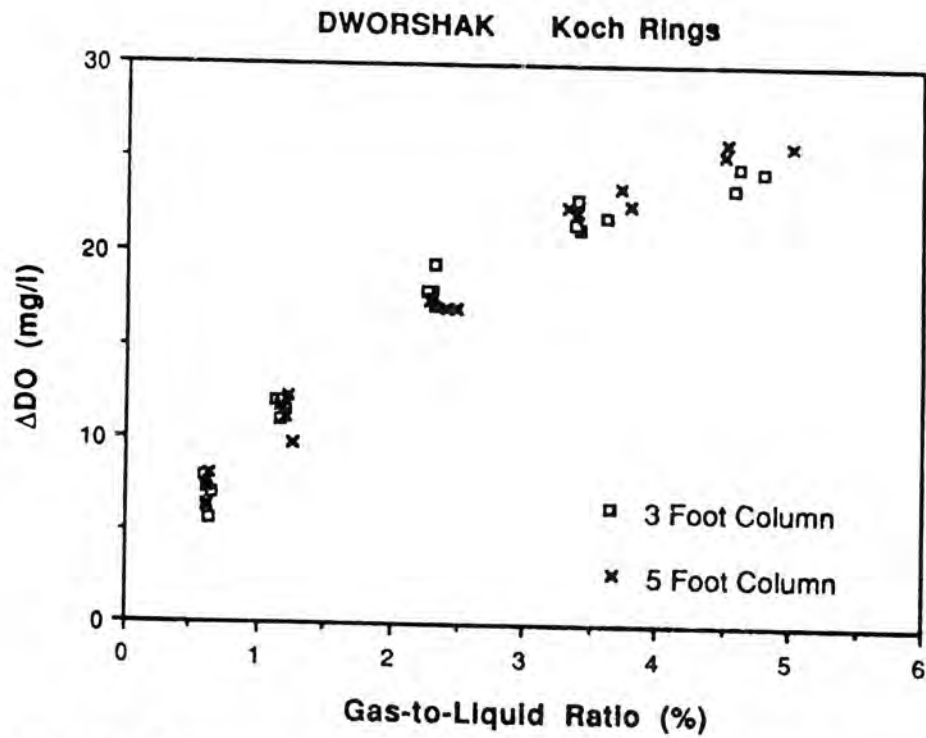


Figure 3. Delta DO or oxygen added (mg/L) to four different water flows through 3 foot and 5 foot, 24-inch sealed columns at gas to liquid ratios ranging from 0.63 to 4.7%.

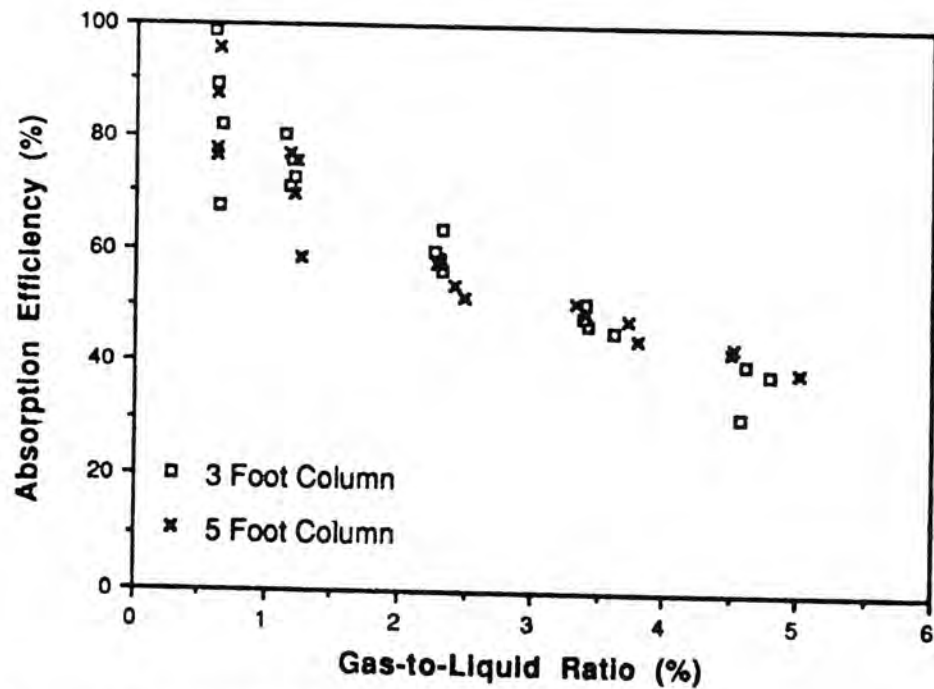


Figure 4. Absorption efficiency (%) at four different water flows through 3 foot and 5 foot, 24-inch sealed columns at gas to liquid ratios ranging from 0.63 to 4.7%.

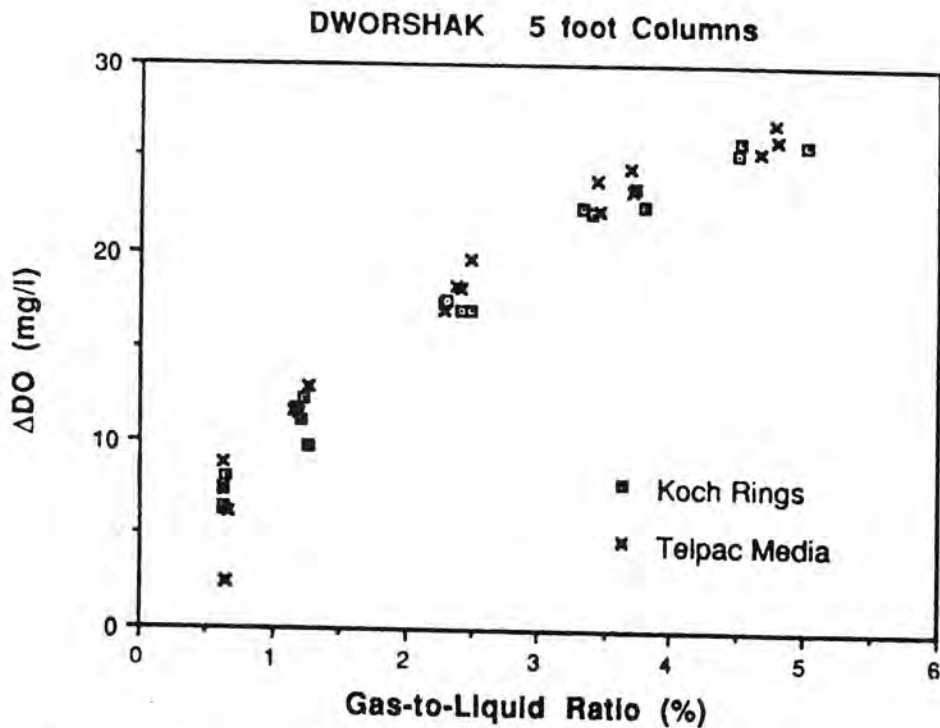


Figure 5. Delta DO or oxygen added (mg/L) to four different water flows through 24-inch sealed columns comparing Koch and Telpack media performance at gas to liquid ratios ranging from 0.63 to 4.7%.

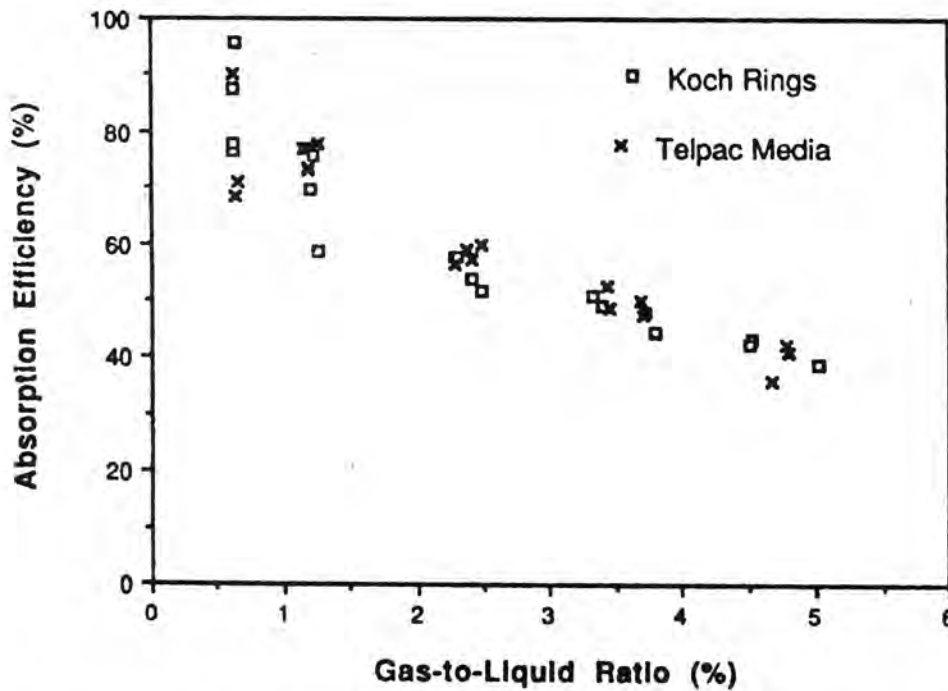


Figure 6. Absorption efficiency (%) at four different water flows through 24-inch sealed columns comparing Koch and Telpack media performance at gas to liquid ratios ranging from 0.63 to 4.7%.

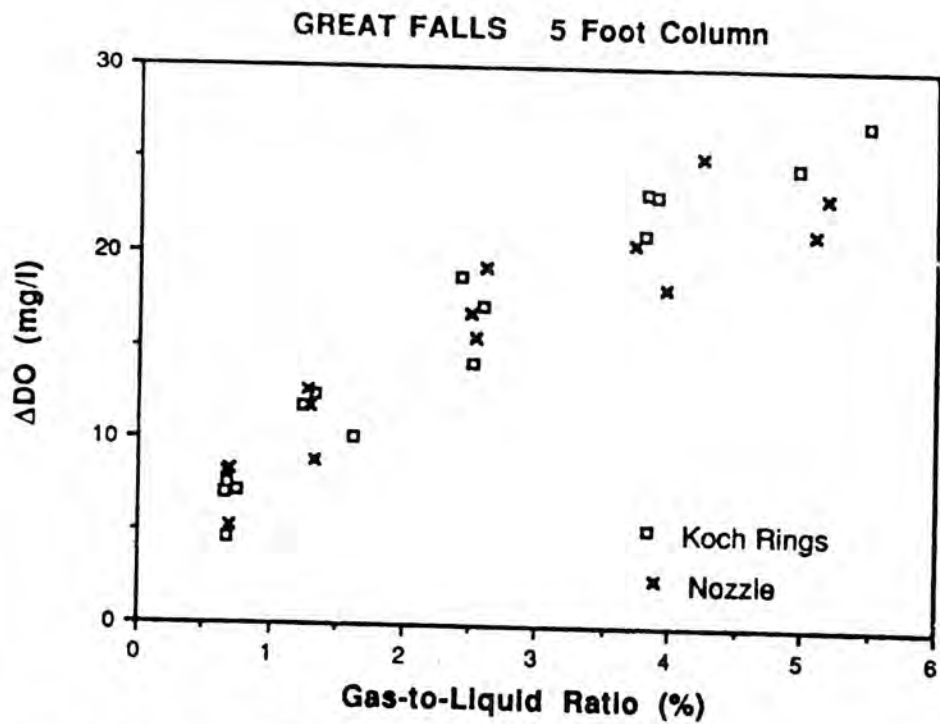


Figure 7. Delta DO or oxygen added (mg/L) to four different water flows through 24-inch sealed columns comparing performance of Koch rings and a spray nozzle mean at gas to liquid ratios ranging from 0.69 to 5.4%.

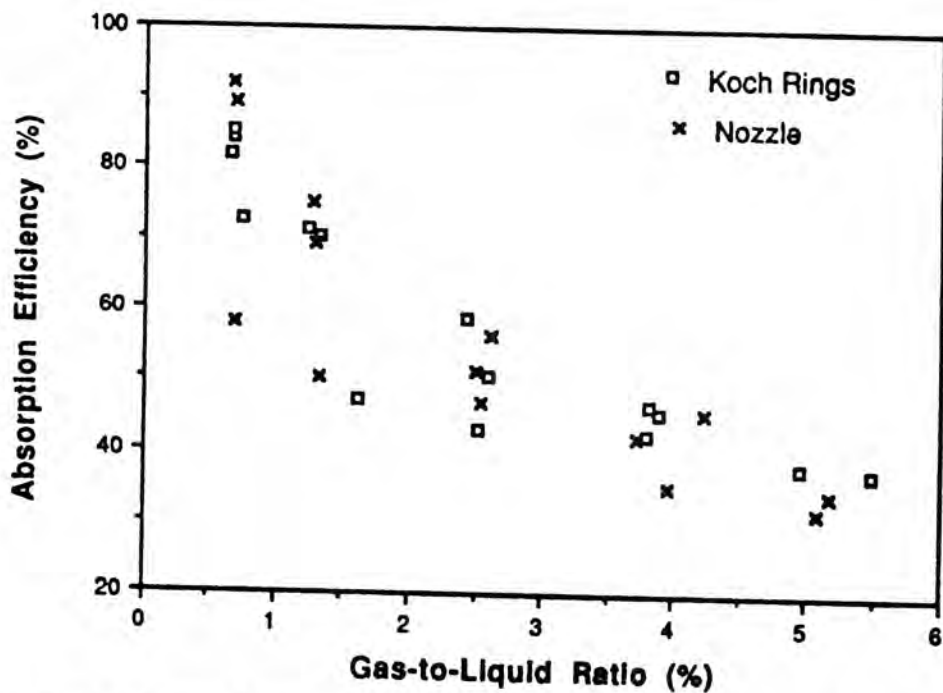


Figure 8. Absorption efficiency (%) at four different water flows through 24-inch sealed columns comparing performance of Koch rings and a spray nozzle at mean gas to liquid ratios ranging from 0.69 to 5.4%.

INJECTED PURE OXYGEN INTO SEALED COLUMNS- MASS TRANSFER MODELING

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Introduction

The Northwest Power Planning Council has established a goal of doubling the size of salmon runs in the Columbia River Basin. The achievement of this important goal is largely dependent upon expanding the production of hatchery fish. The use of pure oxygen has been commonly used to increase carrying capacity of private sector salmonid hatcheries in the Pacific Northwest (Gowan, 1987; Severson et al.,

1987). The use of supplemental oxygen to increase hatchery production is significantly less expensive than the construction of new hatcheries and might save up to \$500 million in construction costs. A better understanding of operation characteristics of pure oxygen absorption systems is needed for planning and design purposes. The development of a mass-transfer model for pure oxygen packed columns is presented in this work.

Mass Transfer Model

The mass transfer model used in this work is based on a two-film theory (Hackney and Colt, 1982):

$$\ln \left| \frac{C^* - C_{in}}{C^* - C_{out}} \right| = \Phi (K_d - KZ) \quad (1)$$

Where

- C* = Saturation gas concentration (mg/l)
- C_{in} = Influent gas concentration (mg/l)
- C_{out} = Effluent gas concentration (mg/l)
- Φ = Relative transfer rate for a specific gas
- K_d = Transfer characteristics for a distribution plate

- K = Transfer characteristics of media (1/ft)
 Z = Packing depth (ft)

A key assumption in this model is that the gas phase is well-mixed and homogeneous throughout the column.

The steady-state effluent gas concentrations (oxygen, nitrogen + argon, and carbon dioxide) from the column are computed using a finite difference approach (Watten et al, In Press) using the following steps:

- (1) Fill the column with oxygen and compute the moles of the gas within the column.
- (2) Set the working composition of gas in the column equal to the influent values.
- (3) Compute the moles of oxygen, nitrogen, and carbon dioxide transferred into the water per time step.
- (4) Compute the moles of gases added to the column from the enriched oxygen flow per time step.
- (5) Re-compute the working composition of gas within the column using 2, 3, & 4.
- (6) Check gas composition for convergence. If solution did not converge, go back to Step 3.

Specific values of the parameters used in this work are:

| | | |
|------------------------------|---|-----------------------------------|
| Media Type | = | 1.5 inch pall rings |
| Void Fraction | = | 94 % |
| K | = | 0.52/ft @ 20 C |
| K _d | = | 0.30 @ 20 C |
| Overall G for O ₂ | = | 2.92 @ 20 C |
| Φ for N ₂ | = | 0.94 |
| Φ for CO ₂ | = | 1.00 |
| Column Height | = | 5.00 ft |
| Column Diameter | = | 2.00 ft |
| Oxygen Purity | = | 100.0 % |
| Hydraulic Loading (low) | = | 86 gpm/ft ² (270 gpm) |
| (high) | = | 245 gpm/ft ² (770 gpm) |

Data Reduction and Reporting

The previously described mass transfer model was used to predict the dissolved gas concentrations of two experimental test conducted at Dworshak National Fish Hatchery. Actual influent dissolved gas concentration, water temperatures, and barometric pressures were used to predict effluent dissolved gas concentrations.

Detailed information on the experimental procedures and results are presented in a companion paper by Dwyer et al. in this proceedings.

Nominal gas flowrates were read off the installed rotameter on the column. This reading was corrected to standard oxygen flowrates (20 C, 1 atm) using the following equation:

$$Q_{O_2} = Q' \left| \frac{1.200}{1.326} \frac{BP+P}{760} \frac{293.15}{273.15+T} \right|^{1/2} \quad (2)$$

where

- Q_{O_2} = Oxygen flow under standard conditions (cubic feet/hour)
- Q' = Equivalent flow of air under local temperature and pressure (cubic feet of air/hour)
- BP = Local barometric pressure (mm Hg)
- P = Effluent pressure (gauge) from rotameter (mm Hg)
- T = Gas temperature at rotameter (C)

The first term in Equation 2 corrects for the difference in molecular weight between air and oxygen. Depending on column pressure and back-pressure in the discharge line to the column, the computed standard oxygen flowrates can range from 80 - 150% of the uncorrected rotameter reading.

Water flowrate was measured using a signet flow meter installed on the influent water line.

The Gas-to-Liquid ratio is expressed as a percent of the water flowrate:

$$G/L (\%) = \frac{Q_{O_2}}{(60Q_{water}/7.48)} \cdot 100 \quad (2)$$

where

$$Q_{water} = \text{water flow (gpm)}$$

The G/L ratio is based on the volumetric flowrate of the actual gas supplied rather than the volumetric flowrate of pure oxygen.

Absorption efficiency (AE) was computed from the oxygen gas flow and change in dissolved oxygen in the column:

$$AE(\%) = \frac{(Q_{water})(3.78)(60)(DO_{out}-DO_{in})(2.205)(10^{-6})}{(Q_{O_2})(0.08309 \text{ lb/ft}^3)(X_{in})} \cdot 100 \quad (3)$$

where

- DO_{out} = Effluent dissolved oxygen (mg/l)
- DO_{in} = Influent dissolved oxygen (mg/l)
- X_{in} = Mole fraction of oxygen gas (assumed to be 0.994)

The density of oxygen under standard conditions is 0.08309 lb/ft³ (Colt and Watten, 1988).

$$\Delta DO = (DO_{out} - DO_{in}) \quad (4)$$

Results

Low Hydraulic Loading- 86 gpm/ft² (270gpm):

The accuracy of the predicted ΔDO and absorption efficiency was good (Figures 1 and 2). The model under-estimated the increase in ΔP (Figure 3), but was better at prediction the effluent nitrogen saturation (Figure 4). The deviation from the observed ΔDO , absorption efficiency, and effluent nitrogen saturation were consistently high for two data points.

High Hydraulic Loading- 245 gpm/ft² (770 gpm):

The results of the model at the high hydraulic loading rate are comparable (Figure 5-8) to the results of the low hydraulic rate.

Discussion

The mass-transfer model was able to accurately predict ΔDO , absorption efficiency, and effluent nitrogen saturation over a wide range of hydraulic loading rates. Problems with the prediction of ΔP are probably related to the use of the measured system pressure at the top of the column as the column pressure. At the high flowrate, the column is definitely flooded (Hackney and Colt, 1982) and the effective pressure within the column is not constant or equal to the value measured above the distribution plate.

This work was based on published column performance data (Hackney and Colt, 1982) using significantly different geometry and flowrates. The predictive ability of this model can be greatly improved by direct determination of key model parameters (K and system pressure) as a function of hydraulic loading.

The change in DO (ΔDO) through the column is equal to:

Acknowledgements

This project was funded in part by the Bonneville Power Administration, the US Fish and Wildlife Service, and the Fish Factory.

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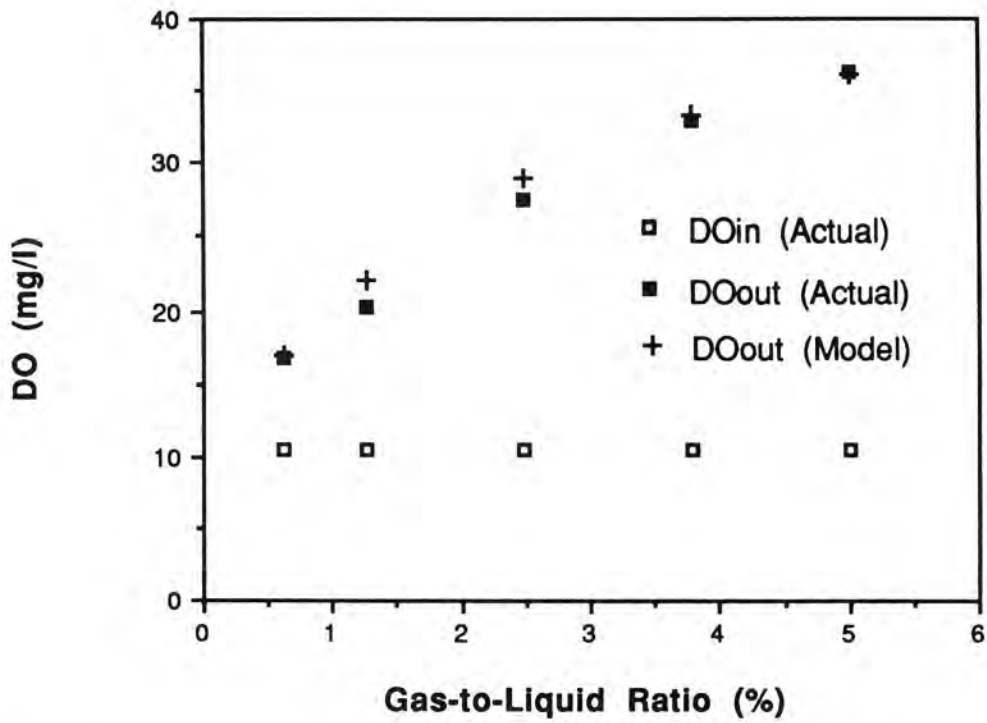


Figure 1 Comparison of Observed and Predicted Change in Dissolved Oxygen (Δ DO) as a Function of Gas-to-Liquid Ratio (270 gpm, 86 gpm/ft²)

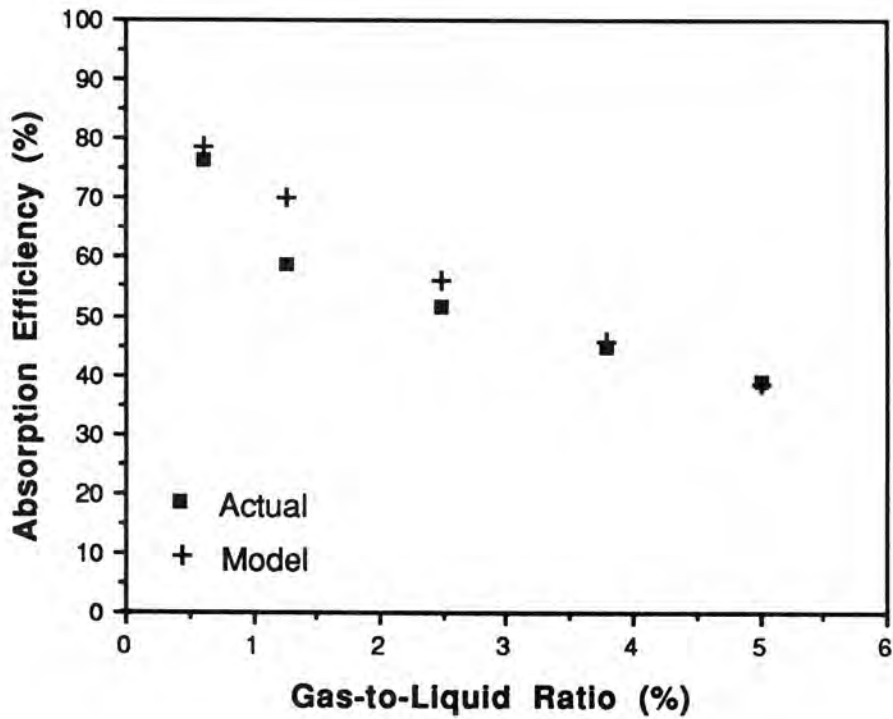


Figure 2 Comparison of Observed and Predicted Absorption Efficiency as a Function of Gas-to-Liquid Ratio (270 gpm, 86 gpm/ft²)

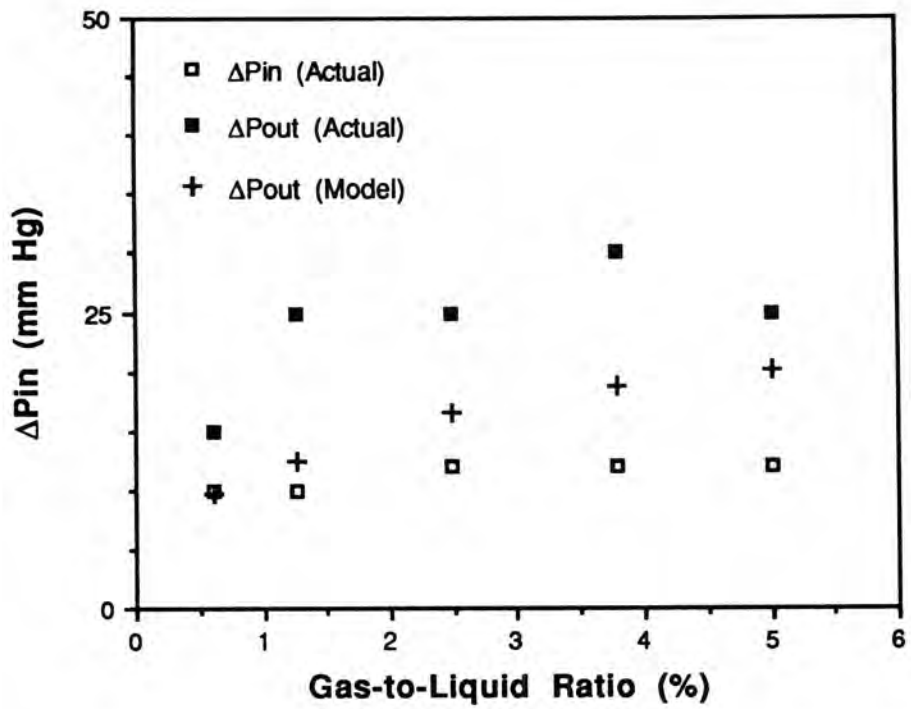


Figure 3 Comparison of Observed and Predicted ΔP as a Function of Gas-to-Liquid Ratio (270 gpm, 86 gpm/ft²)

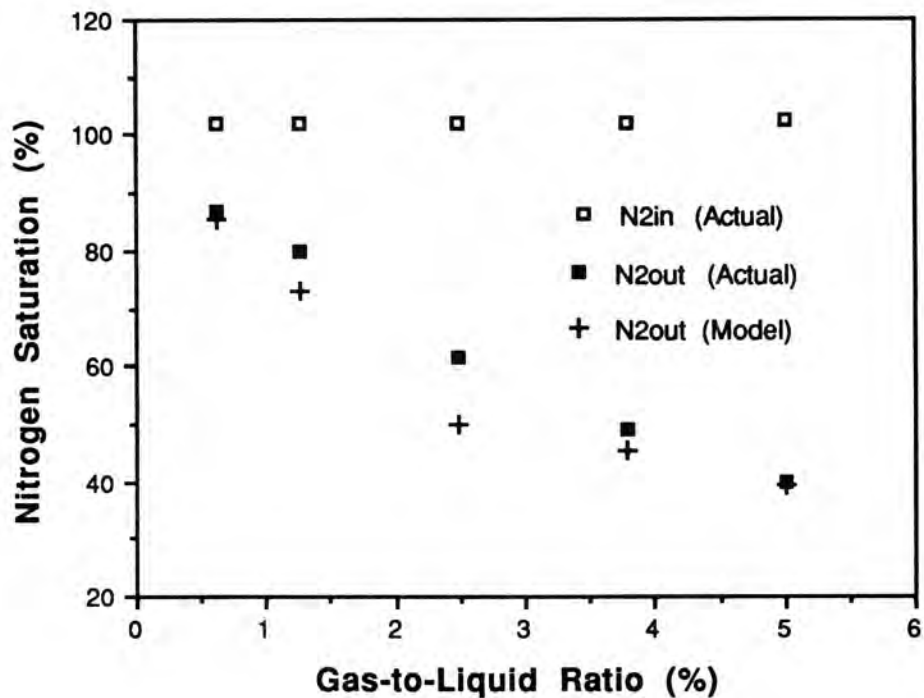


Figure 4 Comparison of Observed and Predicted Nitrogen Saturation as a Function of Gas-to-Liquid Ratio (270 gpm, 86 gpm/ft²)

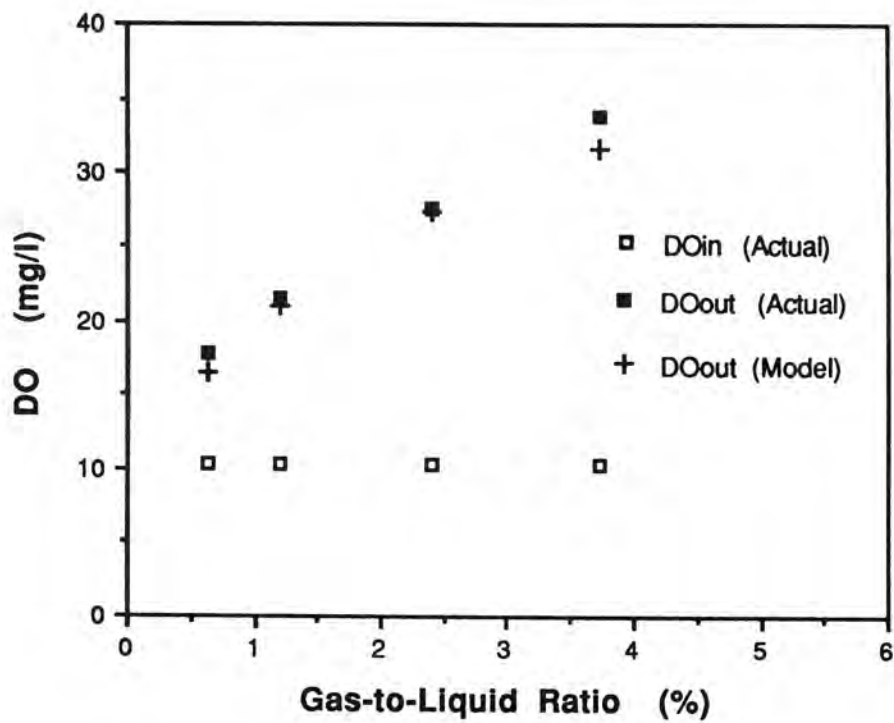


Figure 5 Comparison of Observed and Predicted Change in Dissolved Oxygen (ΔDO) as a Function of Gas-to-Liquid Ratio (770 gpm, 245 gpm/ft²)

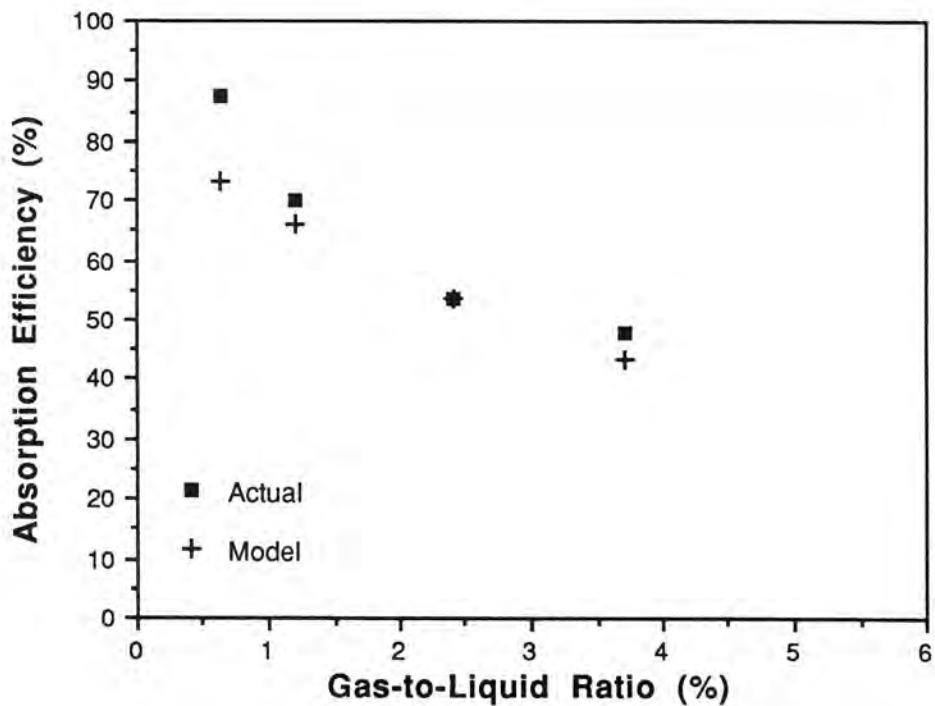


Figure 6 Comparison of Observed and Predicted Absorption Efficiency as a Function of Gas-to-Liquid Ratio (770 gpm, 245 gpm/ft²)

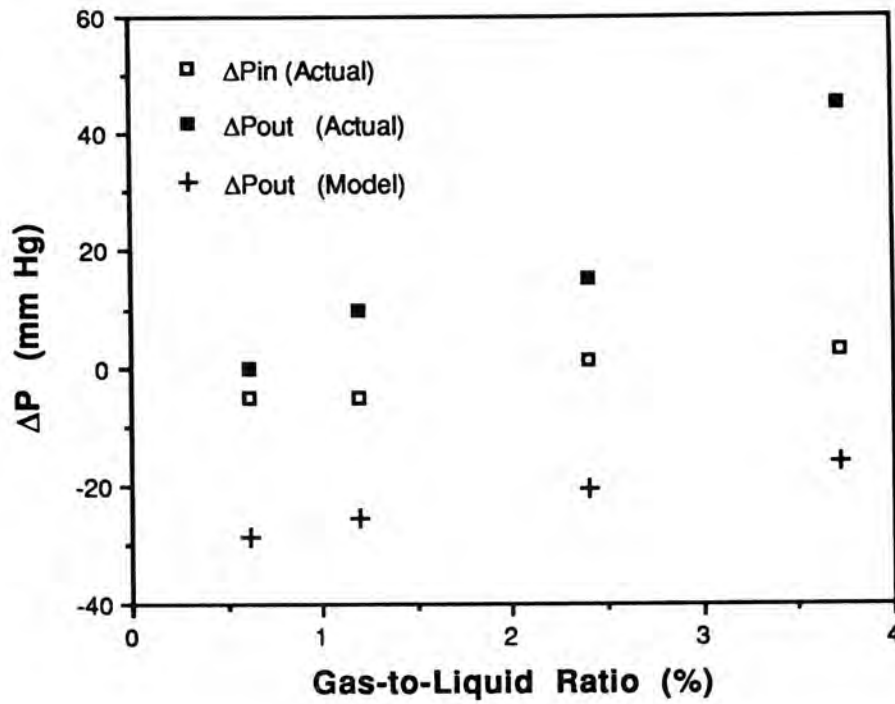


Figure 7 Comparison of Observed and Predicted ΔP as a Function of Gas-to-Liquid Ratio (770 gpm, 245 gpm/ft²)

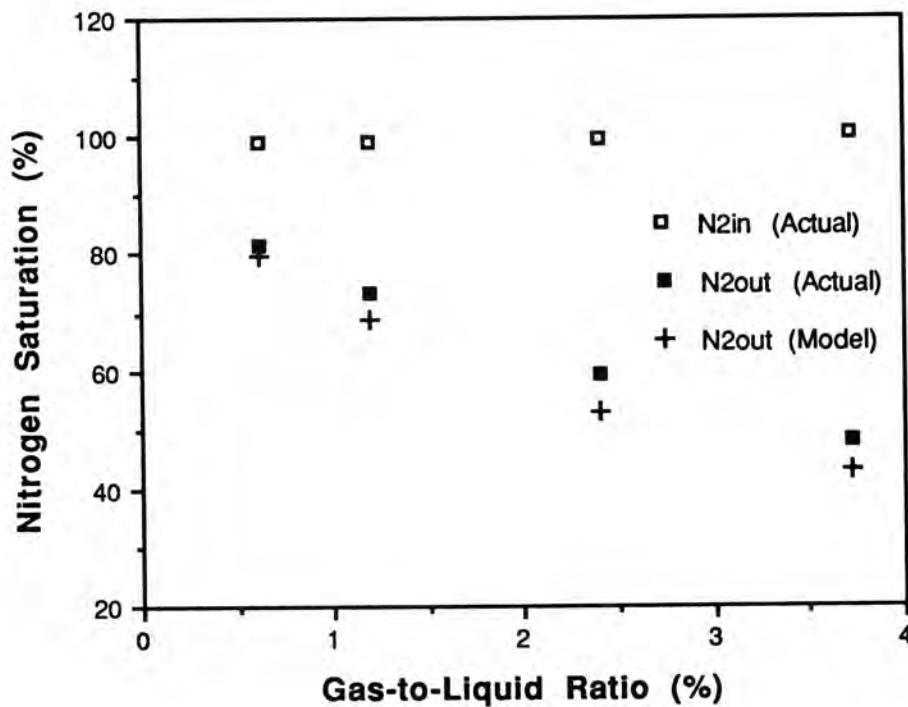


Figure 8 Comparison of Observed and Predicted Nitrogen Saturation as a Function of Gas-to-Liquid Ratio (770 gpm, 245 gpm/ft²)

EFFECT OF DEMAND FEEDERS ON PERFORMANCE OF RAINBOW TROUT REARED IN OXYGEN SUPERSATURATED WATER

by

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ABSTRACT

Demand feeders were used to allow rainbow trout (*Oncorhynchus mykiss*) to feed to satiation while being reared in water supplemented with oxygen to 187% of saturation. There was no improvement in average weight gain or feed conversion efficiency over that in fish reared in untreated water. Hematocrits were slightly lower in fish reared in oxygen supplemented water, but were within the normal range for trout. We concluded that oxygen supplementation does not benefit fish performance unless oxygen saturation in untreated rearing water is lower than about 60%.

INTRODUCTION

The value of oxygen supplementation of hatchery water supplies is well known. Benefits include increased carrying capacity, better water quality, and improved fish health. However, there has been little documentation of the growth performance of fish reared in water supersaturated with oxygen.

Feed costs have been reduced in Michigan state fish hatcheries after oxygen supplementation equipment was installed in the mid-1989's (H. Westers, personal communication). Fish held in single-pass rearing units are exposed to oxygen levels as high as 180% of saturation. However, the effects on growth or feed conversion efficiency have not been documented.

We have previously reared rainbow trout of the Kamloop strain in 180% oxygen supersaturated water for 125 days, but found no significant improvement in weight gain or feed conversion over that in fish reared in water at 90 to 95%

oxygen saturation (Edsall and Smith 1988). Those results agree with production scale studies recently conducted at Saratoga and Jackson (Wyoming) National Fish Hatcheries (NFH), in which rainbow trout and cutthroat trout (*Oncorhynchus clarki*) raised in water supersaturated with 150 to 180% oxygen showed no improvement in growth, feed conversion, or fin quality over fish in control groups (Edsall and Smith 1988; Doulos and Kindschi 1989). However, calculated feed rates, based on a percentage of fish weight, were used to determine the amounts of feed offered in all of these tests. We did not know whether we were limiting growth potential when we assigned fish in the control group and fish in water supplemented with oxygen to a feeding rate that had been designed for subsaturation conditions. The hypothesis in the present study was that if fish were allowed to regulate their own feed intake, or feed to satiation, they would increase feed intake and growth rate when reared in water supersaturated with oxygen.

We repeated the earlier study (Edsall and Smith 1988) except that we trained the fish to use demand feeders and monitored the performance of self-feeding fish held in water supplemented with oxygen. We made the assumption that the fish were feeding to satiation.

METHODS

Rainbow trout of the Arlee strain (mean length 5.5 in) were divided into six lots of 60 fish each and placed into 20 gal rectangular tanks. Three tanks received spring water at 51 degrees F, supersaturated by the injection of pure oxygen into the water supply line. Oxygen was supplied by a model T-2 Nitrox

oxygen generator, in which a molecular sieve is used to separate oxygen from nitrogen. Oxygen from the generator was injected at the top of a polyvinyl- chloride pipe 5 1/2 ft long, 4 in in diameter and packed with 1 in diameter Koch rings. The column was sealed at the top and the bottom extended below the water level. A 1-horsepower electric water pump was used to raise water to the top of the column. The water and oxygen were mixed in the column, and supersaturated water was then collected in a headbox and distributed to each tank. The three control tanks also received spring water at 51 degrees F, but it was untreated.

We used small electronic demand feeders (Cablegation Controls, Hagerman, ID) that supply feed when the fish pulls on a target suspended in the water. An arm lifts the target out of the water and at the same time turns a small conveyor belt forward about 1/4 in, dropping several pellets of feed. A counter on the box records the number of times the feeder is activated.

The fish learned to use the feeders within a few days. During the 100-day study, we weighed the fish every 2 weeks, and recorded the feed added to the feeders. Hematocrits were determined for 10 fish from each tank at the end of the trial.

Dissolved oxygen, nitrogen, and total gas pressure were determined daily. We measured the hyperbaric pressure of gas in the water with a Weiss satumeter (Eco Enterprises, Seattle, WA) and dissolved oxygen and temperature with a meter manufactured by the Yellow Springs Instruments Co., Inc. (Yellow Springs, OH). Nitrogen and total gas were calculated by using formulas described by Colt (1984).

The average saturation of the oxygen supplemented water was $186.7 \pm 7.8\%$ ($x \pm SD$) oxygen and $78.5 \pm 2.1\%$ nitrogen; total gas pressure was $101.2 \pm 0.4\%$. The average gas saturations of the water that held the control group was $95.4 \pm 2.3\%$ oxygen and $100 \pm 1.0\%$ nitrogen; total gas pressure was $99.0 \pm 1.0\%$.

The treatment means for average weight gain, feed conversion, and hematocrit were

analyzed by a general linear models analysis of variance (Hintze 1987); individual tanks of fish were the experimental units. The level of significance was accepted as being $p < 0.05$.

RESULTS AND DISCUSSION

There were no differences in average weight gain or feed conversion between rainbow trout reared in oxygen supersaturated water and those reared in untreated water (Table 1).

Hematocrits were lower in fish from the oxygen supplemented group than in control fish. (The hematocrits gives an indication of the oxygen-carrying capacity of the blood, and fish reared in an oxygen enriched environment would have less need for oxygen transport capability). However, the hematocrits we recorded were well within the normal range for trout (Miller et. al. 1983; Korcock et. al. 1988), and post-rearing survival studies (Edsall and Smith 1988) have shown that fish reared at high oxygen saturations are not at a disadvantage when they are returned to a normal environment.

Results of this study indicated that fish fed to satiation and reared in oxygen supersaturated water did not outperform fish reared in water slightly below oxygen saturation. These results agree with those from earlier laboratory and production scale studies with fish not fed to satiation (Edsall and Smith 1988; Doulos and Kindschi 1990). Brett (1979) concluded from the work of Chiba (1966), Stewart et. al. (1967), and Herrmann et. al. (1963) that an oxygen concentration of about 5 mg/L is critical for growth and acceptable feed conversion efficiency; at lower concentrations, both factors are proportionally suppressed as oxygen concentration decreases. At oxygen concentrations above the critical zone, growth and feed conversion efficiency are completely independent of oxygen concentration. Westers (1981) observed that the performance of fish reared in water substantially below oxygen saturation (<60%) was adversely affected, and that oxygen supplementation increased growth rate. However, fish normally

reared in water nearer to oxygen saturation (>90%) probably would not improve in performance when reared in water supplemented with oxygen, if other environmental factors such as fish density, water temperature, and ammonia concentration remained unchanged.

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Table 1. Performance of rainbow to (initial average weight, 31 g) fed to satiation in oxygen supersaturated water for 100 days (all values are means of three replicates.)^a

| Performance factor | Oxygen saturation (%) | |
|-------------------------------------|-----------------------|------|
| | 187 | 95 |
| Weight gain (g per fish) | 82.0 | 61.0 |
| Feed conversion factor ^b | 1.18 | 1.13 |
| Hematocrit (%) | 37.3 | 40.8 |
| Survival (%) | 99.4 | 98.9 |

^a Among the pairs of values, only the hematocrits differed significantly ($P < 0.05$). Pooled standard errors of the mean for weight gain, feed conversion factor, hematocrit, and survival were 8.13, 0.05, 0.92, and 0.88 respectively.

^b Weight of feed added to demand reeders/weight gained.

DEVELOPMENT OF A DECISION SUPPORT SYSTEM FOR SALMON HATCHERY MANAGEMENT

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INTRODUCTION

Oregon State University, in cooperation with the Bonneville Power Administration, has begun a research project aimed at developing a comprehensive, user-friendly mathematical model for simulating, and eventually optimizing, management strategies for salmonid hatchery production. Started in October, 1989, the project is expected to last three years, with the resulting model to enter a testing phase during 1991. The model will contain detailed mathematical descriptions of fish growth, feeding and metabolism, water quality (e.g. DO, ammonia, pH, solids), water transport and treatment systems, raceway and rearing unit dynamics, and energy and resource consumption of a facility.

The model will have two simulation modes: 1) *batch* and 2) *interactive*. In *batch* mode, the model will be given a set of production goals (e.g. fish release number, weight and dates) and facility specifications. The model will then simulate the response of each raceway and each lot of fish in the hatchery, reporting state variable responses, flagging potentially limiting areas in the production scheme (e.g. low DO in a raceway), and generating appropriate feeding strategies and fish management schedules to reach the stated production goals in a near-optimal manner. In the *interactive* mode, the simulation steps through the operation of a hatchery at a user-defined time interval, typically one day. At the end of each simulated time interval, the model stops and generates a series of reports describing the state of all hatchery components, again flagging potential trouble areas, and makes recommendations for possible remedial measures. Any or all of these reports can be viewed. Additionally, the user may at this time alter the value of any of the system state variables (e.g. if data has been collected from a raceway and the predicted state needs to be

corrected), generate a consultative report describing the overall state of the hatchery with recommendations for appropriate management strategies to deal with any problems, and perform any of a number of management actions (e.g. splitting lots, controlling aerators, altering feed schedules, etc.). The model then proceeds to simulate the response of the hatchery system for the next time interval, again providing the user an opportunity to interact with the system at the end of the specified interval. In this manner, the user of the system can quickly explore the effects of specific management strategies on the system's response in a highly interactive environment, and use the system to monitor day-to-day activities within the hatchery.

RATIONALE

This research project was originally conceived to address concerns about hatchery effectiveness and productivity, and to provide a research tool for understanding the basic biological and ecological interactions occurring within the hatchery environment. A large body of knowledge has been generated regarding salmonid hatchery production. Because these are complex systems, it is difficult to synthesize this knowledge and utilize it to generate appropriate management strategies. Mathematical models provide a tool for capturing, manipulating and maintaining this knowledge within a well-defined framework to provide improved management capabilities for optimizing facility scheduling and resource utilization, and predicting how the system will respond to various dynamic influences. The process of constructing such a model is useful to identify areas where knowledge is lacking; similarly, this process can be used to prioritize research to "fill in the holes" in the current knowledge base. Once a model is built and validated, it can be useful for playing "what-if" games to determine how various

management strategies might influence production costs and effectiveness. A validated model may also be used, through sensitivity analysis, to determine how sensitive the system is to changes in each of the inputs, allowing researchers and managers to focus on those variables which have the greatest influence on particular hatchery components.

PROJECT OBJECTIVES

- 1) Evaluate existing computer simulation models of salmonid allowing prioritization of future research efforts.
- 2) Expand, refine, validate and document these models to incorporate a comprehensive, state of the art representation of hatchery operations suitable for addressing critical areas of hatchery management.
- 3) Identify critical areas of research and develop guidelines for an integrated program of hatchery research.
- 4) Implement algorithms for optimizing hatchery operations and maximizing production and/or economic returns.

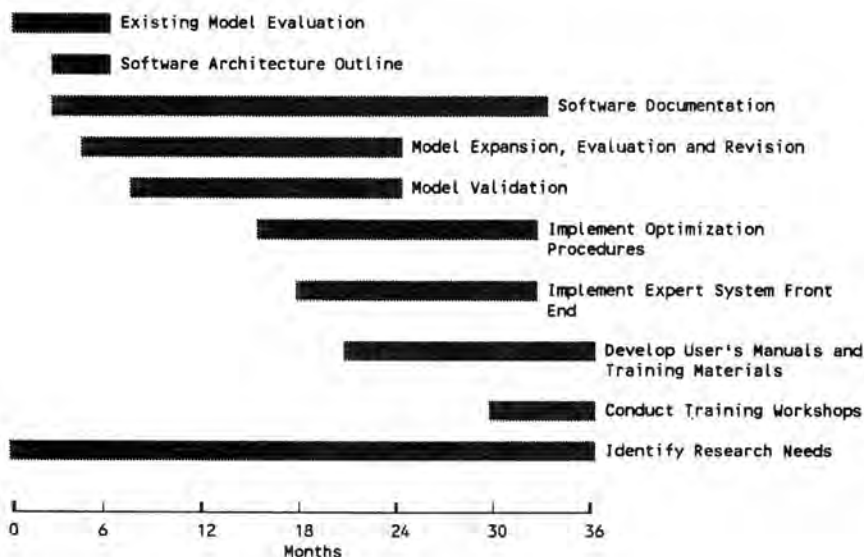
- 5) Develop a "user-friendly" environment for model use.

BENEFITS

A number of benefits to the hatchery industry will be realized as a result of this research, including:

- 1) Improvement of the efficiency of salmonid hatchery production facilities through simulation of alternative management strategies and strategy optimization.
- 2) Development of a repository for capturing, clarifying and enhancing existing knowledge hatchery biological/physical processes and interactions in a readily accessible framework.
- 3) Identification of areas where gaps in our understanding of "systems" level processes exist, allowing prioritization of future research efforts.
- 4) Identification of areas, through sensitivity analysis, where improvement and optimization will provide the greatest benefits.
- 5) Establishment of a framework for incorporating and utilizing new knowledge as this knowledge becomes available.

Timeline for Implementation



BETTER INFORMATION FOR HATCHERY MANAGERS- A DATABASE METHOD FOR TRANSFERRING INFORMATION TO THE HATCHERY AND FROM THE HATCHERY

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Introduction

Better information for hatchery managers in information that is more consistent, and more accessible than with traditional record keeping methods. Information stored in database format can meet current needs while minimizing redundancy and improving consistency, and can easily be retrieved at a future date. Use of common database files can also serve as an efficient medium of exchange among offices.

However, the use of database methodology throughout an organization requires more cooperation than other record keeping methods. As stated in previous papers and presentations, "The basic premise of this work is that fish hatcheries and fisheries managers have common data needs and are working toward the same goals. By working together, the amount of effort expended for data collection can be kept to a minimum, and data quality can be enhanced."

Two previous papers have described some of the basic elements of a database system for documenting various activities at U.S. Fish and Wildlife operated fish hatcheries and fisheries assistance offices in the lower Columbia River Basin. These files document the number of adults returning to the hatchery, the number of adults spawned, the number of eggs taken, and their disposition through initial feeding. All distribution of fish from the hatchery is also documented in data files.

A description of data collection utilizing a portable computer will be followed by a discussion of data returned to hatchery managers.

Adult Sampling Utilizing A Portable Computer

The sampling of returning adults provides information on the success of various age

classes, and more detailed information through recovery of coded wire tags. Length, sex, marks, and identification needed to link fish heads with length and sex data are usually recorded on data sheets at the spawning site. These data are usually referred to as biodata. Later, data are then entered into an electronic format so that they can be analyzed by appropriate computer programs. Data entry is time consuming and error prone.

Portable microcomputers capable of running Database Management Systems (DBMS), and storing large amounts of data are now available for use in the field. One such unit was utilized during fall chinook sampling at Spring Creek NFH. A clear plastic bag was used to protect the computer from water and blood. A data entry program which creates two copies of the data (one on the hard drive and one on a floppy disk) was used.

After retrieval and reading, coded wire tag information is added to the files. Age information is added as scales are read. Since all needed data are typed directly into data files, the process of transferring data from paper to electronic media is eliminated.

Resistance to Change Nearly Universal

It seems intuitively obvious that the elimination of an entire step in a process would save time and therefore be beneficial. It would therefore seem reasonable to assume acceptance of this new procedure.

Nevertheless implementation of this change elicited demands that all data be printed out so that ages could be written on paper, then transferred from the paper into the data files. This demand was accompanied by an upheld pencil along with the remark, "There is nothing wrong with using paper and pencil." Although this type of reaction may be expected from someone with

computer phobia, it came from a recently educated, computer literate individual.

Returning Information to the Hatchery Manager

Information in biodata files is analyzed by a suite of programs known collectively as Age COmposition programs.

It is traditional to send printed output of analyzed data to hatchery managers. However, the Vancouver office processed approximately 1,500 heads, and read over 8,000 scales. Analysis of this data produced a stack of paper about 1 1/2" thick. Duplication of hardcopy is time consuming.

It is possible to send data files and the programs which produce printed output on one floppy disk. Hatchery managers can produce hardcopy simply by running the required programs. Although the processing of data requires computer time at the hatchery, additional expense is not incurred.

Although these programs work primarily with biodata, they also make use of information in data files originally created by the hatchery, and used for other purposes.

For example, after determining the number and percentage of each age group in the

sampled fish, the total number of fish which returned to the hatchery is obtained from the Fish Removal file created by the hatchery. The program then calculates the number of fish in each age class for the total run. All of this information is printed on a Hatchery Return Data Form given both to the hatchery manager and other agencies. Since the hatchery uses the Fish Removal file to document daily spawning, removal of fish "dead in pond", etc., during the spawning season, the number of fish on the Hatchery Return Data Form will agree with this information.

Returning coded wire tag information is obtained from biodata files. The Age Composition program uses information in files containing release information to calculate and print the percent return for coded wire tags retrieved. Release information is placed in data files at the hatchery when fish are released. Reports are generated by software using these data files. This same information is available years later, both at the hatchery, and at other offices, when percent return is calculated.

The repeated use of data recorded only one time accounts for some of the increased consistency observed when data base methods are used. Since data are recorded in a stable electronic format, they can easily be retrieved by computer programs.

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| Olson | Craig | NWIFC | 6730 Martin Way E. | Olympia | WA | 98506 |
| Olson | Ron | NWIFC | 6730 Martin Way | Olympia | WA | 98506 |
| Olson | Todd | CEDC Fisheries | 250 - 36th Street | Astoria | OR | 97103 |
| Olson | Wayne | Dworshak NFH | P.O. Box 18 | Ahsahka | ID | 83520 |
| Oman | Leni | WDW | 600 North Capitol Way | Olympia | WA | 98504 |
| Orendorff | John | Bioproducts, Inc. | P.O. Box 429 | Warrenton | OR | 97146 |
| Orr | Wesley | Ennis NFH | 180 Fish Hatchery Road | Ennis | MT | 59729 |
| Orthmann | R.A. | Rt. 1 Box 42 | | LaCenter | WA | 98629 |
| Orwicz | Kris | Aquatic Biotics | 1245 Cunningham | Dixon | CA | 95620 |
| Osborne | Gary | Rocky Reach Hatchery | Rt. 3 Box 3136 | E. Wenatchee | WA | 98802 |
| Owsley | D.E. | Dworshak NFH | P.O. Box 18 | Ahsahka | ID | 83520 |
| Page | David | WDF | 15702 52nd Street East | Sumner | WA | 98390 |
| Paiya | Mike | USFWS- Warm Springs NFH | P.O. Box 790 | Warm Springs | OR | 97761 |
| Palmer | Ted | Common Sensing Inc. | 7595 Finch Road, N.E. | Bainbridge I. | WA | 98110 |
| Parke | Clyde | Allison Creek Brood Trout | Station, Box 394 | Coleman | ALTA | TOK 0M0 |
| Parrish | Dave | Oregon Pacific Salmon | 23154 U.S. Highway 101 N. | Brookings | OR | 97415 |
| Pastor | Steve | USFWS | 9317 N.E. 99th, Suite 1 | Vancouver | WA | 98682 |
| Paulsen | Robert R. | WDF | 4203 Central Park Drive | Aberdeen | WA | 98520 |
| Pearson | Karen | Global Aqua-UAS, Inc. | 355 Ericksen Avenue ST. 421 | Bainbridge I. | WA | 98110 |
| Pegg | Marty | Valdez Fisheries Devel. Assn. | P.O. Box 125 | Valdez | AK | 99686 |
| Pellissier | Rene' | Prince William Sound Aqua | P.O. Box 1110 | Cordova | AK | 99574 |
| Penn | Dean | Quileute Fisheries Dept. | P.O.Box 187 | La Push | WA | 98350 |
| Pennell | William | Malaspina College | 1239 Okanagan Place | Nanaimo | B.C. | V9R 5Z5 |
| Penny | John | Turtle Rock | Rocky Reach Dam | Wenatchee | WA | 98801 |
| Peterson | Don | B.C. Ministry Of Environment | 780 Blanshard Street | Victoria | B.C. | V8V 1X5 |

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|--------------|------------|------------------------------|------------------------------|--------------|------|---------|
| Phillips | Patrick E. | 2008 Pacific Place | | Mt. Vernon | WA | 98273 |
| Phillips | Ray | Biosponge Aqua Products, Co. | 23 E. Brundage, PO Box 2089 | Sheridan | WY | 82801 |
| Phillipson | Ken | NWIFC | 6730 Martin Way East | Olympia | WA | 98506 |
| Poe | Sidney D. | Silverado Fisheries Base | CDF&G, P.O. Box 47 | Yountville | CA | 94599 |
| Popochock | Denis | Minter Creek Hatchery | 12710 124th Ave. Ct. K; P.N. | Gig Harbor | WA | 98335 |
| Pratschner | Greg | USFWS, Leavenworth NFH | P.O. Box 549 | Leavenworth | WA | 98826 |
| Presseau | Roland | IBEC Aquaculture Corporation | P.O. Box 789 | Port McNeill | B.C. | VON 2R0 |
| Prestegard | Eric | Prince William Sound Aqua | P.O. Box 1110 | Cordova | AK | 99574 |
| Preston | Paul | Nanaimo River S.E.P. | 271 Pine Street | Nanaimo | B.C. | V9R 2B7 |
| Radford | Linda | Darrah Springs Hatchery | P.O. Box 8 | Paynes Creek | CA | 96075 |
| Raistakka | Wesley I. | Colman NFH | Rt. 1 Box 2105 | Anderson | CA | 96007 |
| Rasmussen | Ulf | South Tacoma Hatchery | 7723 Phillips Road SW | Tacoma | WA | 98498 |
| Reece | Mark L. | Humtulpils Hatchery | P.O. Box 129 | Humtulpils | WA | 98552 |
| Rhine | Dean | Valdez Fisheries Development | Box 125 | Valdez | AK | 99686 |
| Rieben | David | Big Creek Hatchery | Rt. 4, Box 594 | Astoria | OR | 97103 |
| Ringenbach | Roger | Faivre of America | 833 9th Street | Santa Monica | CA | 90403 |
| Robards | Steve | Chelan Hatchery | HCR Box 52 | Chelan | WA | 98816 |
| Roberts | Russel | P.O. Box 34 | | Mt. Vernon | WA | 98273 |
| Roberts | Steve | WOW | 3860 Chelan Highway | Wenatchee | WA | 98801 |
| Robinette | Karen | Prince William Sound Aqua | P.O. Box 670 | Whittier | AK | 99693 |
| Rockhold | Anne | USFWS | 342 Whitman | Leavenworth | WA | 98826 |
| Rogers | Dave | ODFW | 43863 Greer Dr. | Leaburg | OR | 97489 |
| Rogers | Dick | Hood Canal | P.O. Box 606 | Hoodsport | WA | 98548 |
| Rogers | Robert | Washington Dept of Fisheries | W191 Deyette Rd | Shelton | WA | 98584 |
| Rogers | Tom | Sawtooth Fish Hatchery | HC 64, Box 9905 | Stanley | ID | 83278 |
| Roley | Dennis | Bioproducts, Inc. | P.O. Box 429 | Warrenton | OR | 97146 |
| Rosholt | Thane | 2019 28th Ave. W. | | Seattle | WA | 98199 |
| Rossmann | Steven P. | 2425 S.E. 92nd | | Portland | OR | 97216 |
| Rowan | Jan | USFWS | P.O. Box 80 | Neilton | WA | 98566 |
| Rowland | Rick A. | ODF&W | 2917 S.E. 142nd Place | Portland | OR | 97236 |
| Rueschmann | Peter | Waring Marine | 3113 - 2nd Avenue | Port Alberni | B.C. | V9Y 4C4 |
| Russum | Jerry | Elk River Hatchery | 95163 Elk River Road | Port Orford | OR | 97465 |
| Rutledge | Ray | Enertech/Rutledge Envir. | 17508 48th St. S.E. | Snohomish | WA | 09290 |
| Sampson | Don | P.O. Box 638 | | Pendleton | OR | 97801 |
| Schaeffer | Leslie | ODF&W | 17330 SE Evelyn St. | Clackamas | OR | 97015 |
| Schamber | Tim | Fall Creek Hatchery | 2418 East Fall Creek Road | Alsea | OR | 97324 |
| Schneider | Richard | Clear Springs Trout Company | P.O. Box 712 | Buhl | ID | 83341 |
| Schroder | Larry | Skagit Hatchery | 5937 Fish Hatchery Lane | Marblemount | WA | 98267 |
| Schultz | Murray | ODF&W | P.O. Box 50 | Portland | OR | 97207 |
| Schwab | Rick | Rangen Inc. | P.O. Box 706 | Buhl | ID | 83316 |
| Sciankowy | Craig | DFO | 8301 Mahonia St. | Mission | B.C. | V2V 6E5 |
| Scott | Marilyn | WDF | 23 Scott Rd | Glenwood | WA | 98619 |
| Scribner | Tom | Yakima Indian Tribe | P.O. Box 151 | Toppenish | WA | 98948 |
| Semen | Doug | MariSource (FAL/Heath) | P.O. Box 9037 | Tacoma | WA | 98409 |
| Shake | Bill | USFWS | 911 N.E. 11th Ave. | Portland | OR | 97232 |
| Shanahan | Bea | Argent Laboratories, Inc. | 8702 152 Avenue, N.E. | Redmond | WA | 98052 |
| Sheldon | Ray | ODF&W | 17330 S.E. Evelyn Street | Clackamas | OR | 97015 |
| Sheldrake | T.J. | | 8135 S.W. Berryhill Rd. | Beaverton | OR | 97005 |
| Shippentower | Gene E. | 16047 E. Burnside St. | | Portland | OR | 97233 |
| Siaz | Eileen | Darrah Springs Hatchery | P.O. Box 8 | Paynes Creek | CA | 96075 |
| Siemens | Laird | BC Fisheries Branch | Kootenay Trout Hatchery | Wardner | B.C. | VOB 2J0 |
| Silvey | Brian | 69787 E. Barlow Trail Rd. | | Rhododendron | OR | 97049 |
| Smith | Charlie | Bozeman Fish Technology Ctr | 4050 Bridger Canyon Rd | Bozeman | MT | 59715 |
| Smith | Quentin | Klaskanine Hatchery | Rt. 1, Box 764 | Astoria | OR | 97103 |
| Smith | R.Z. | NMFS- ETSD | 911 N.E. 11th, Suite 620 | Portland | OR | 97232 |
| Smith | Shawn W. | CDF&G | 1600 Hatchery Road | Arcata | CA | 95221 |
| Solazzi | Mario | ODFW | 850 SW 15th | Corvallis | OR | 97331 |
| Sorensen | Dan | Makah NFH | P.O. Box 730 | Neah Bay | WA | 98357 |
| Sparrow | Hugh | 1655 Warren Gardens | | Victoria | B.C. | V8S 1S9 |
| Stanley | Charlie | Cedar Creek Hatchery | 33465 Highway 22 | Hebo | OR | 97122 |
| Steele | Earl | Bellingham Voc-Tech Inst. | 3028 Lindbergh Ave. | Bellingham | WA | 98225 |
| Stewart | Bruce | NWIFC | 6730 Martin Way E. | Olympia | WA | 98506 |
| Stickell | Trent | ODF&W | P.O. Box 59 | Portland | OR | 97207 |
| Stone | Wayne | U.S.F.W.S | Larson Road | Underwood | WA | 98651 |
| Streufert | Jon | USFWS | Dworshak NFH | Ahsahka | ID | 83520 |
| Swann | Trace | 2401 E Street | | Bellingham | WA | 98225 |
| Tansley | Bill | ODF&W | P.O. Box 130 | Camp Sherman | OR | 97730 |
| Taylor | Gib | USFWS | 1221 Ebb Tide Terrace | Olympia | WA | 98502 |

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|-----------|------------|-------------------------------|-----------------------------|----------------|------|---------|
| Thorson | William | Entiat NFH | 6970 Hatchery Drive | Entiat | WA | 98822 |
| Tipping | Jack | WDF | 2101 Hwy 508 | Onalaska | WA | 98570 |
| Todd | Neil L. | Diversified Ova-Tech Ltd | Box 237 | Merritt | B.C. | VOK 280 |
| Turner | Lynda | McKernan Hatchery | W. 411 Deyette Road | Shelton | WA | 98584 |
| Uplinger | Richard M. | CDF&G | 1664 Hatchery Road | Arcata | CA | 95521 |
| Van Ree | Gary | Global Aqua-USA, Inc. | 355 Ericksen Avenue St. 421 | Bainbridge I. | WA | 98110 |
| Vendshus | Sharon | ODFW | 17220 S.E. Evelyn | Clackamas | OR | 97015 |
| Vincent | David | Malaspina College Aquaculture | 200 5th Street | Nanaimo | B.C. | V9R 5K2 |
| Volkhardt | G.C. | NWIFC | 6730 Martin Way E. | Olympia | WA | 98506 |
| Voskuilen | Reece | Sverdrup Corporation | P.O. Box 369 | Bellevue | WA | 98009 |
| Vreeland | Robert | NMFS, 7600 Sand Pt. Way NE | BIN C15700, Bldg. 1 | Seattle | WA | 98115 |
| Wagner | Paul | Pyramid Lake Fisheries | Star Route | Sutcliffe | NV | 89510 |
| Wall | Gary G. | ADF&G | P.O. Box 5-337 | Ft. Richardson | AK | 99505 |
| Wallien | William | Winthrop NFH | P.O. Box 429 | Winthrop | WA | 98826 |
| Walters | Tim | 2549-B SW Pickford | | Corvallis | OR | 97333 |
| Ward | Larry | Lower Elwah Hatchery | 1674 Lower Elwah Rd. | Port Angeles | WA | 98362 |
| Warren | Jim W. | USFWS | 911 N.E. 11th Ave. | Portland | OR | 97232 |
| Warren | Roger | ODFW | 580 Fish Lane Road | Butte Falls | OR | 97522 |
| Westby | Carl | Pacific Biological Station | Hammond Bay Road | Nanaimo | B.C. | V9R 5K6 |
| White | Dan | Northwest Scale Systems | 2100 W. Broadway #5 | Eugene | OR | 97402 |
| White | Lorne | ADF&G- FRED Division | 211 Mission Road | Kodiak | AK | 99615 |
| Wilson | Wendy | Biosponge Aquaculture Co. | 23 East Brundage, Box 2089 | Sheridan | WY | 82801 |
| Winters | Randy | ODF&W | Star Route Box 73 | Idanda | OR | 97350 |
| Wirth | Rob | 2950 N.E. 23rd | Apt. 12 | Gresham | OR | 97030 |
| Wold | Einar | 26507 N.E. 10th Ave. | | Vancouver | WA | 98642 |
| Wolff | Klaus | Ministry Of Environment | R.R. 1, Site 11 | Summerland | B.C. | VOH 120 |
| Wong | Ron | USFWS- Makah NFH | P.O. Box 730 | Neah Bay | WA | 98357 |
| Woodard | Robert D. | 2950 N.E. 23rd #20 | | Gresham | OR | 97030 |
| Woody | Stanley | Beaver Creek Hatchery | 28 Beaver Creek Road | Cathlamet | WA | 98612 |
| Wright | Terry | NWIFC | 6730 Martin Way E. | Olympia | WA | 98506 |
| Yaskovic | John | ODFW | P.O. Box 59 | Portland | OR | 97207 |
| Yeager | Gary | Nehalem Hatchery | Rt. 1, Box 292 | Nehalem | OR | 97131 |
| Zimmerman | Brian | Paradise Bay Seafarms | P.O. Box 1540 | Port Townsend | WA | 98368 |

NWFCC Mailing List For Commercial Exhibitors

| Company Name | Address | City/State | Zip Code |
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| Argent Laboratories | 8702-152nd Avenue, NE | Redmond, Wa. | 98052 |
| Bill Nelson Co. | 1549 N.W. 49th | Seattle, Wa | 98117 |
| Bio-Sponge Aqua Products, Co | 23 E. Brundage, PO Box 2089 | Sheridan, Wy | 82801 |
| Bioproducts, Inc. | P.O. Box 429 | Warrenton, Or. | 97146 |
| Common Sensing Inc. | 7595 Finch Road, NE | Bainbridge Is. Wa | 98110 |
| Enertech | 17508 48th St. SE | Snohomish Wa. | 98290 |
| Engineered Products | P.O. Box 30 | Philomath, Or | 97370 |
| Faivre of America | 833 9th Street | Santa Monica, Ca | 90403 |
| Industrial Plastics, Inc. | 740 South 28th Street | Washougal, Wa | 98671 |
| J.L. Eager Inc. | P.O. Box 476 | N. Salt Lake, Ut | 84054 |
| Jensorter, Inc. | 20225 Harvest Lane | Bend, Or. | 97701 |
| Lummi Fisheries Supply, Inc. | 851 Coho Way | Bellingham, Wa | 98225 |
| Marisource (FAL Heath tray) | 4540 S. Adams, PO Box 9037 | Tacoma, Wa | 98409 |
| Moore-Clark Co. Inc. | P.O. Box M | LaConner, Wa | 98257 |
| Northwest Scale Systems | 2100 W. Broadway, Ste. 5 | Eugene, Or. | 97402 |
| Ozone Tech./Dawson Controls | P.O. Box 30222 | Portland, Or | 97230 |
| P.R.A. Manufacturing Ltd. | P.O. Box 774, Station A | Nanaimo, B.C. | V9R 5M2 |
| Protect A Cover | 2424 Manor Way | Everett, Wa | 98204 |
| Reiff Fiberglass Company | Route 4, Box 183 | Walla Walla, Wa | 99362 |
| Silver Cup Fish Feed | P.O. Box 155 | Manzanita, Or. | 97130 |
| Waring Marine | 3113 - 2nd Avenue | Port Alberni, B.C. | V9Y 4C4 |

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| Text Book of Fish Health by Dr. George Post | Argent Chemical Labs. | Ron Morinaka | BPA |
| 100x Bi-scope | Argent Chemical Labs. | Vance McGowan | Anadromous, Inc. |
| Woman's Float Jacket | Lumni Fisheries Supply | John Larson | BC Fisheries Branch |
| Men's Float Jacket | Lumni Fisheries Supply | John Ennor | DIPAC |
| Gerber Knife | Bioproducts Inc. | Ole Jan Flatraker | Global Aqua-USA |
| Gerber Knife | Bioproducts Inc. | Jon Streufert | USFWS |
| Gerber Knife | Bioproducts Inc. | Carl Westby | D.F.O. Canada |
| Buck Knife | Bioproducts Inc. | Dan Herrig | USFWS |
| Buck Knife | Bioproducts Inc. | Ed Labiske | ODFW |
| Fish Measuring Board | Marisource (FAL Heath Tray) | Christine Hansen | USFWS |
| Room Package for 2 | Salishan Lodge | Jeff Drongesen | OSU Dept. Micro. |
| Room Package for 2 | Salishan Lodge | Gary Osborne | WDF |
| Room Package for 2 | Salishan Lodge | Bill Hutchinson | IDFG |
| Room Package for 2 | Salishan Lodge | Dave Owsley | USFWS |
| Quart Thermos | Jensorter | Bob Molony | WDF |
| Gift Package of Food | Murray Elevators | Ron Wong | USFWS |
| Insulated Tote Bag | Bill Nelson Co. | Bill Shake | USFWS |
| Alpine Vineyards Wine | Engineered Products | Art Filliger | Clearwater Waste Systems, Inc. |
| Ceramic Salmon Plaque | Moore Clark Co. | Jerry Fisher | ODFW |
| Ceramic Salmon Plaque | Moore Clark Co. | Tony Novotony | BIOMED Research Labs |
| \$50 Gift Certificate | J.L. Eagar Inc. | David Fast | Yakima Indian Nation |
| Fish Counter | Faivre of America | CDFG | Calif. Fish. & Game |