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BERNARD MICHAEL KEPSHIRE, JR. for the MASTER OF SCIENCE
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William McNeil

Juvenile fall chinook salmon were acclimated to full strength sea water at 66, 73, and 80 days after hatching. Survival in sea water for 14 days was used as the criterion for successful acclimation.

The growth of salmon acclimated to sea water was slower over the period of observation (134 days) than the growth of fish from the same parental stock held in fresh water and 50 percent sea water at similar temperatures. The additional energy required for osmoregulation in full-strength sea water was thought to reduce growth.

The larger size of fish held in fresh water and 50 percent sea water was reflected in their higher stamina as determined by their ability to swim against a current of known velocity. Where fish acclimated to sea water were similar in size, though older in age, to fish held in fresh and low salinity water, there were no obvious differences in stamina.

From the results of this work, it seems feasible to acclimate

juvenile fall chinook salmon to full-strength sea water within 90 days after hatching. Low mortality can be anticipated from exposure to high salinity, but growth will be slowed at least temporarily. Successful acclimation can be achieved by placing fish in 50 percent sea water 40 days after hatching and feeding them for 50 days before transferring them to full-strength sea water.

Growth and Stamina of Sea-Water Acclimated Juvenile Fall Chinook Salmon

by

Bernard Michael Kepshire, Jr.

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Professor of Fisheries
in charge of major

Redacted for Privacy

Head of Department of Fisheries and Wildlife

Redacted for Privacy

Dean of Graduate School

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GROWTH AND STAMINA OF SEA WATER ACCLIMATED JUVENILE FALL CHINOOK SALMON

INTRODUCTION

Water pollution, dams, logging, and other activities of man have decimated salmon populations in many streams and rivers (U.S. Department of Health, Education, and Welfare, 1961; Bell, DeLacy and Paulik, 1967; Haas, 1965). Fresh water hatcheries have mitigated some losses and now sustain a substantial portion of salmon production in the Pacific Northwest (Cleaver, 1969). The possibility of acclimating certain species of Pacific salmon, especially chinook salmon, Oncorhynchus tshawytscha (Walbaum, 1792), to sea water early in life could potentially reduce costs of hatcheries and provide new avenues for maintaining and enhancing salmon stocks.

The broad objective of this study is to explore opportunities for producing fall chinook salmon which are fully capable of oceanic existence early in life. Should young fish survive the oceanic environment and feed on the copious food supply, there is a possibility that they may return to spawn at an earlier age than salmon reared for longer periods in fresh water. Selective breeding of returning adults which had been acclimated to sea water as very young fish might produce a population of chinook salmon which are physiologically capable of placement into the ocean soon after yolk absorption.

Early acclimation of fall chinook salmon to sea water (30‰) has been demonstrated in the laboratory by exposing fish to gradual increments in salinity after the vitelline vessicle had been absorbed. Wagner, Conte and Fessler (1969) found that chinook salmon could tolerate 50 to 67 percent sea water (15‰ - 20‰) as alevins soon after hatching. In another study, Black (1951) found that young coho salmon, O. kisutch (Walbaum, 1792), could be adapted quite readily to 50 percent sea water as indicated by their ability to regulate blood chloride.

To estimate the potential survival of young salmon acclimated to sea water after their release from hatcheries, both growth and stamina should be evaluated. Fast growth of juvenile salmon has been observed in 50 percent sea water. Coho salmon initially weighing 0.5-1.0 grams exhibited better growth in 40 and 60 percent sea water (12‰ - 18‰) than in fresh water (Canagaratnam, 1959). One-year-old chinook salmon also exhibited better growth in 50 percent sea water (17.7‰) than in fresh water (Bullivant, 1961). Bullivant found, however, that chinook salmon raised in full-strength sea water (35.4‰), after spending eight days in 50 percent sea water, grew much more slowly than fish remaining in fresh water.

Stamina tests have indicated that diseased or otherwise unhealthy fish perform poorly in swimming tests when compared to healthy fish (Thomas, Burrows and Chenoweth, 1964). There have been no studies, to my knowledge, where stamina of fish acclimated to salt water has been compared to fish in fresh water.

The present study compares the growth and stamina of young fall chinook salmon which have been acclimated to sea water with that of fish reared in fresh water and in 50 percent sea water. This research was financed by the National Science Foundation's Office of Sea Grant Programs. Fall chinook salmon eggs were supplied by the Fish Commission of Oregon. Rearing tanks and fresh and sea water distribution systems used in this study at the Port Orford Marine Research Laboratory were constructed with funds from Curry County, Oregon.

GENERAL METHODS

Experiments reported in this thesis involved two major groups of fall chinook salmon which shall be referred to as Groups I and II. Both groups were obtained as eyed eggs from the Fish Commission of Oregon, Elk River Hatchery. Salmon were selected from each group for exposure to sea water at varying ages and through varying sequences of increasing salinity.

Four 100-gallon plywood tanks were used to contain the test salinities. The tanks were coated with fiberglass and were mounted on a frame (Figure 1). Two one-inch diameter Polyvinyl Chloride (PVC) ball valves mounted above the uppermost tank were adjusted to allow equal volumes of fresh and ocean water to mix and produce 50 percent sea water (16‰). Two half-inch diameter PVC ball valves entered a second tank and were adjusted to permit equal volumes of ocean water and 50 percent sea water from the uppermost tank to mix and provide 75 percent sea water (24‰). Likewise, equal flows of fresh water and 50 percent sea water were allowed to mix in a third tank to provide 25 percent sea water (8‰). Finally, an equal volume of fresh water and 25 percent sea water were allowed to mix in a fourth tank to produce 12.5 percent sea water (4‰). Short plastic nozzles from valves controlling water entering each tank were in juxtaposition to facilitate mixing.

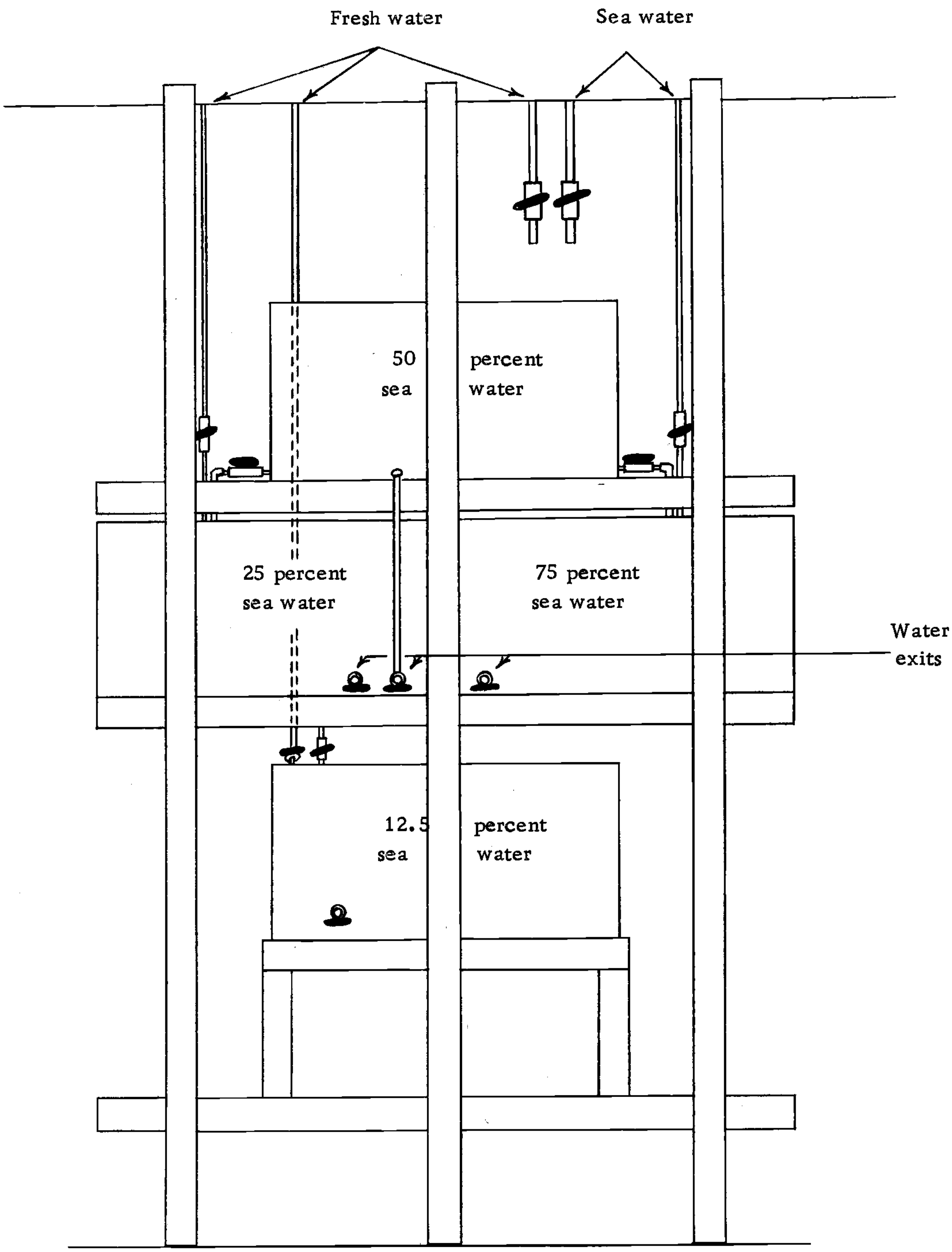

Figure 1. Apparatus for obtaining various dilutions of sea water.

The tank with 12.5 percent sea water had one exit line, but each of the other three tanks had two exit lines on opposite tank sides. All exit lines were controlled by half-inch diameter PVC ball valves.

Eggs and alevins were held initially in shallow, screened trays which were plumbed to receive fresh water. The fry were later transferred to 100-gallon rearing tanks after initiation of feeding. The rearing tanks were plumbed to receive 25, 50, and 75 percent sea water from the sea-water dilution apparatus, and fresh water and 100 percent sea water from separate lines.

Tanks for rearing juveniles after initiation of feeding were constructed of plywood and coated inside with fiberglass. Water entrance and exit lines were located at opposite ends of the tanks (Figure 2). The overflow water was collected in a "V"-shaped trough and discarded. Water was added at the rate of one-half gallon per minute (gpm).

Procedures with Group I Salmon

Eggs were hatched on a fine mesh nylon screen suspended in a 200-gallon tank. An adequate oxygen supply was insured by water upwelling through the screen and overflowing through a pipe placed above the screen. Eggs and alevins were shielded from light until most of the fry began to swim up. Dead eggs and alevins were removed periodically with a squeeze-bulb apparatus.

The following steps were followed to acclimate the Group I

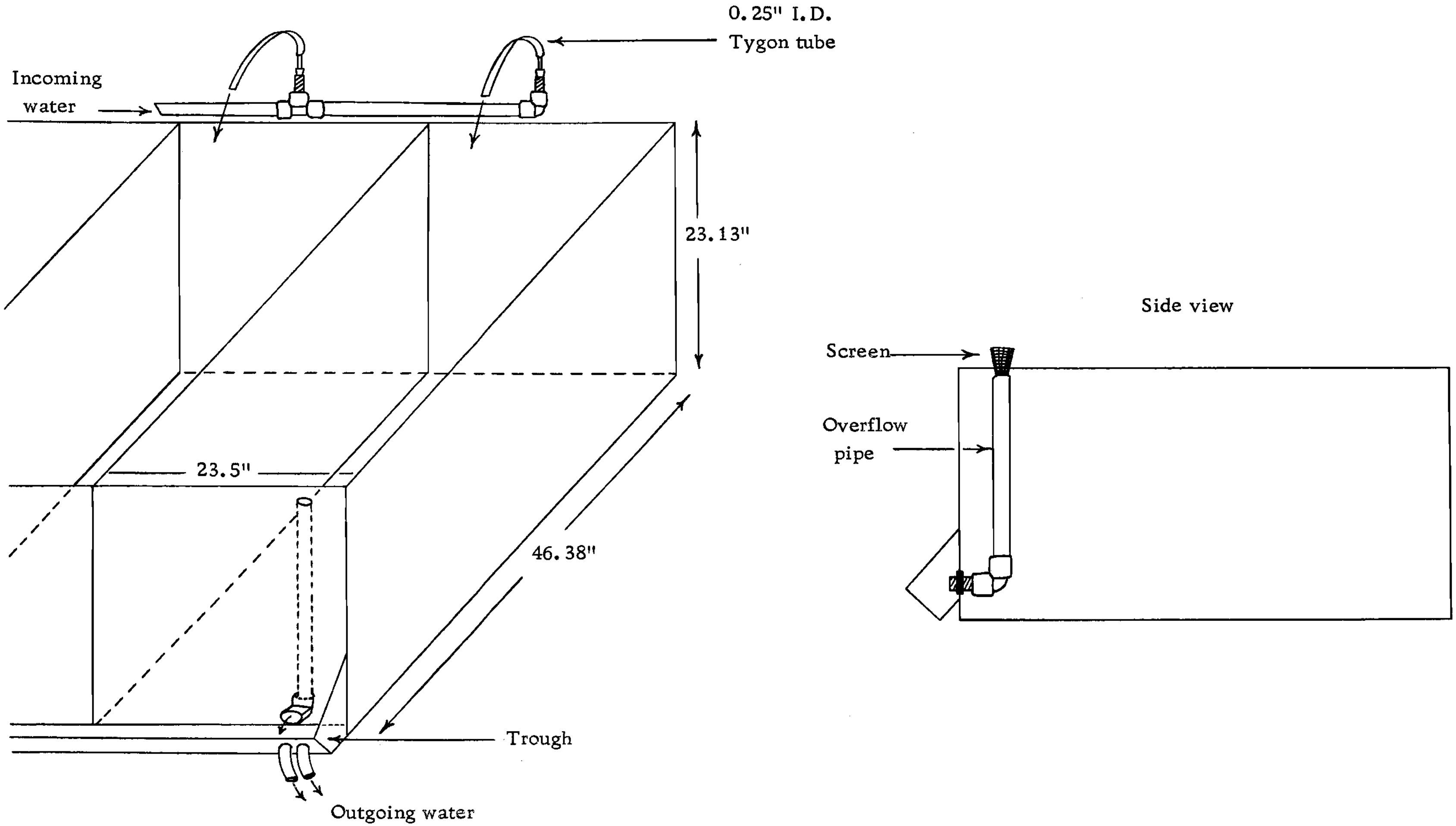

Figure 2. A typical 100-gallon rearing tank.

salmon to sea water:

(1) Forty-seven days after hatching, 1200 fish were removed from the hatching tank and divided into six subgroups of 200 fish each. Two subgroups were placed in separate rearing tanks receiving 25 percent sea water[1/]; two subgroups were placed in separate tanks receiving 50 percent sea water; one subgroup was placed in a tank receiving 75 percent sea water; and one subgroup was placed in a tank receiving fresh water.

(2) Sixty-six days after hatching, one subgroup which had been in 50 percent sea water and one subgroup which had been in 25 percent sea water for 19 days were transferred to separate tanks receiving 75 percent sea water. One subgroup which had been in 25 percent sea water (47 days after hatching) was transferred to a tank receiving 50 percent sea water.

(3) Eighty days after hatching, a subgroup in 50 percent sea water and two subgroups which earlier had been transferred to 75 percent sea water (66 days after hatching) were all transferred to 100 percent sea water. At this time, a subgroup which had been in 75 percent sea water (47 days after hatching) was transferred to 100 percent sea water.

[1/] Since the laboratory receives a constant supply of fresh sea water, sea water dilution percentages experienced minor fluctuations in ‰ as a function of the sea water salinity during the day.

The sequences of exposure to increased salinity are depicted for the subgroups in Table 1.

Table 1. Sequence of acclimation of Group I salmon.

	Subgroups were placed in water of known salinity 1/ at the following ages: 2/			
Subgroup	0 days	47 days	66 days	80 days ⟶ 146 days
Ia	0%			
Ib	0% ⟶	25% ⟶	50% 3/	
Ic	0% ⟶	50% ⟶	⟶	100%
Id	0% ⟶	75% ⟶	⟶	100%
Ie	0% ⟶	50% ⟶	75% ⟶	100%
If	0% ⟶	25% ⟶	75% ⟶	100%

1/ Salinity is expressed as a percentage of full-strength sea water.

2/ Age is expressed as days after hatching.

3/ No arrow after a percent salinity indicates that the subgroup remained in this salinity for the remainder of the experimental period which terminated at 146 days of age.

Procedures with Group II Salmon

Eggs were hatched on a nylon screen suspended in a 200-gallon tank.

Shallow plastic pans were used for alevins and fry in Group II. A fine mesh screen stapled to a removeable wooden frame was placed in each pan. Water volume above the screen, where fish resided, was approximately 1.2 gallons. Water entered each pan from the bottom through a half-inch I. D. pipe and exited through holes just below the top of the pan (Figure 3). Water was added at the rate of one-half gpm.

The following steps were followed to acclimate the Group II salmon to sea water:

(1) Eighteen days after hatching, nine subgroups of 300 alevins each were exposed to varying sea-water dilutions and to fresh water. Two of the subgroups were placed in separate fresh-water pans; three were placed in 12.5 percent sea water; three were placed in 25 percent sea water; and one was placed in 50 percent sea water.

(2) Thirty-two days after hatching, a tenth subgroup consisting of 100 alevins was taken from fresh water and placed in 50 percent sea water. This subgroup was discarded two weeks later, being of no further experimental use.

(3) Thirty-six days after hatching, the three subgroups receiving 12.5 and three receiving 25 percent sea water were placed in 50 percent sea water.

(4) Forty-one days after hatching, the eight subgroups of fish (those in fresh and 50 percent sea water) were transferred to 100-gallon rearing tanks with no change in salinity.

(5) Fifty-four days after hatching, four subgroups in 50 percent sea water were exposed to 75 percent sea water.

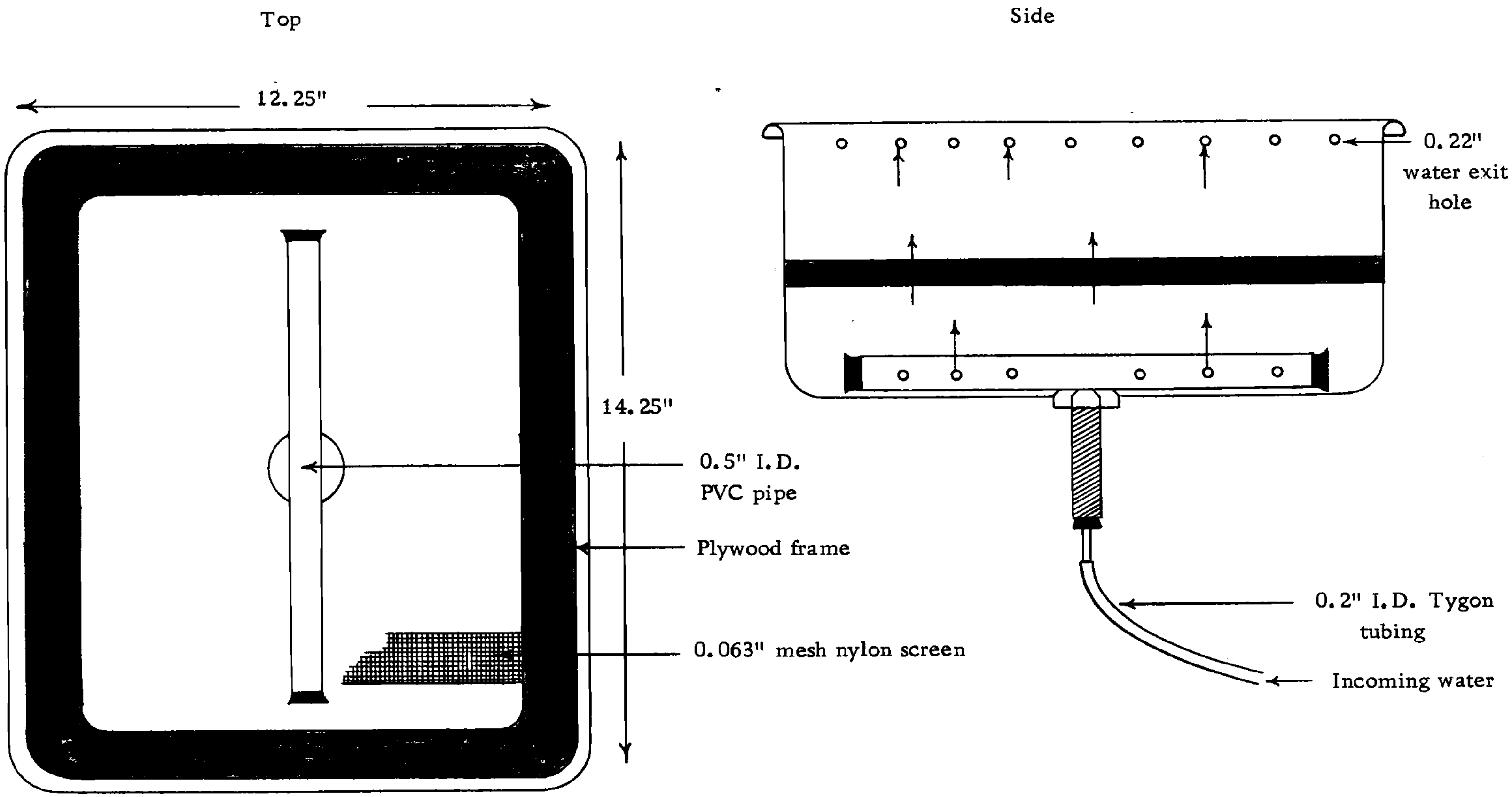

Figure 3. A two-gallon rearing pan for alevins and fry.

(6) Sixty-six days after hatching, all salmon in 75 percent sea water were exposed to 100 percent sea water.

(7) Seventy-three days after hatching, a subgroup which had been in 25 percent sea water 18 days after hatching and in 50 percent sea water 36 days after hatching was exposed to 100 percent sea water.

The sequences of acclimation of the different subgroups are summarized in Table 2.

Table 2. Sequence of acclimation of Group II salmon.

	Subgroups were placed in water of known salinity at the following ages:						
Subgroup	0 days	18 days	32 days	36 days	54 days	66 days	73 days → 117 days
IIa	0%						
IIb	0%						
IIc	0%	12.5%		50%	75%	100%	
IId	0%	12.5%		50%	75%	100%	
IIe	0%	12.5%		50%			
IIf	0%	25%		50%	75%	100%	
IIg	0%	25%		50%	75%	100%	
IIh	0%	25%		50%			100%
IIi	0%	50%					
IIj	0%		50%				

Each change to a higher salinity was accomplished by substituting water of a higher salinity for water of a lower salinity. Approximately eight hours was required to complete a change to higher salinity.

For both major groups, a constant water volume/number of fish ratio was maintained in the rearing tanks to keep constant any density dependent factors, such as oxygen concentration and waste metabolites.

BEHAVIOR AND SURVIVAL

My discussion on behavior and survival will first consider observations on Group I salmon, to be followed by a discussion of observations on Group II salmon. I shall make frequent reference to the subgroups listed in Tables 1 and 2.

Observations on Group I Salmon

Group I salmon (subgroups Ib, Ic, Ie, and If) transferred as fry from fresh water to 25 and 50 percent sea water 47 days after hatching exhibited little or no stress; mortality remained low and no unusual behavior was observed.

Salmon transferred from fresh water to 75 percent (subgroup Id) sea water, on the other hand, exhibited unusual behavior in comparison to fish which remained in fresh water (subgroup Ia) or which had been transferred to 25 and 50 percent sea water. Fish in 75 percent sea water stopped feeding at the water surface within 24 hours after exposure to high salinity, although some of them continued to take sinking food. They were also easily frightened and tended to remain near the bottom of the tank; whereas, fry in fresh water and in 25 and 50 percent sea water remained distributed throughout their tanks. No deaths occurred in 75 percent sea water until the fifth day after initial exposure, at which time about five percent of the original fish died.

No further mortalities occurred until the eighth day, but for the succeeding ten days the total mortality was rather high (20 percent).[2/] After this period of high mortality, the salmon surviving in 75 percent sea water appeared to be able to tolerate their hypertonic environment.

Sixty-six days after hatching, subgroups in 25 and 50 percent sea water were transferred to 75 percent sea water (subgroups Ie and If). These fish suffered no apparent ill effects in the higher salinity, as indicated by low mortality and normal feeding behavior. All salmon had completed absorption of their yolk within 60 days after hatching.

A few days prior to the placement of subgroups Ic, Id, Ie, and If in 100 percent sea water, some fish had jumped out of their tanks and others had escaped through the overflow pipes. The situation was remedied by screening all overflow pipes and by placing removable screens over the tanks at night. Also, a board was placed across one end of each tank to provide cover for fish wishing to hide.

All Group I salmon, regardless of their acclimation route, exhibited the same changed behavior when placed in 100 percent sea water. Compared to salmon in fresh water and in 50 percent sea water, those in 100 percent sea water were easily frightened, and,

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[2/] All mortalities are expressed as a percentage of dead fish out of the total number of fish originally placed in a tank.

except for a few individuals, ceased feeding for 24 to 48 hours after initial exposure to 100 percent sea water. The tendency for salmon acclimated to full-strength sea water to exhibit fright reactions continued throughout the experiment. This behavior is discussed further in the section on growth.

Some mortality occurred within the first two days after fish were placed in 100 percent sea water, but further deaths did not occur until after the salmon had lived in sea water for seven days or longer. Mortality rate generally did not exceed one fish every two days up to the end of the observation period except when fish were starved for 54 hours in connection with stamina testing. This will be discussed in more detail later.

Mortality of Group I salmon is summarized in Table 3.

It was assumed that surviving salmon had adapted to 100 percent sea water within ten days after initial exposure (Wagner, Conte and Fessler, 1969), so the mortality which was observed after ten days of exposure was suspected to have partly been caused by disease. Some dead and dying salmon were taken to the Marine Science Center, Newport, for more detailed examination, but no evidence of disease was found.

Dying fish exhibited a very dark skin color and abnormal behavior. All became sluggish and a few would lose body equilibrium and appear to gulp air at the water surface prior to death. A few of

Table 3. Mortality of Group I salmon.

Age range during which the stated mortality occurred	Mortality in 75% sea water	Mortality during initial shock following exposure to 100% sea water	Mortality in 100% seawater after 54 hours without food[1/]	Mortality in 100% sea water for the remainder of the experimental period	Total mortality
	47-80	81-83	130-131	82-129 131-146[2/]	
Subgroup					
Ia	—	—	—	—	1.0%
Ib	—[3/]	—	—	—	0
Ic	—	4.5%	0	1.5%	6.0%
Id	22.5%	0.5%	0.5%	2.0%	25.5%
Ie	0	0.5%	1.5%	4.0%	6.0%
If	0	3.0%	3.5%	5.0%	11.5%

[1/] All subgroups were starved for 54 hours prior to endurance testing using a Category I test schedule; mortality occurred within 24 hours after feeding was resumed.

[2/] These age ranges account for all mortality in 100 percent sea water not accounted for during the period of initial shock (81-83 days) or after 54 hours without food (130-131 days).

[3/] Indicates that the heading does not pertain to the subgroup.

the fish had bloated, mucus-filled abdominal cavities. The possibility that these conditions resulted from acute osmotic stress will be discussed later.

Observations on Group II Salmon

Some Group II salmon were placed in 100 percent sea water as early as 66 days after hatching (subgroups IIc, IId, IIf, and IIg); whereas, the earliest that Group I salmon were placed in 100 percent sea water was 80 days.

Alevins placed in 12.5 (subgroups IIc, IId, and IIe) and 25 percent (subgroups IIf, IIg, and IIh) sea water 18 days after hatching appeared to be more active and to begin feeding earlier than fish held in fresh water. All fish placed in 50 percent sea water (subgroup IIi) 18 days after hatching died within five to 12 days.

Although 300 alevins were placed in each rearing pan, some escaped by jumping out or by swimming through the water exit holes. The number of fry remaining in each rearing pan at the time of transfer to the tanks ranged from 116 to 258.

To determine the age that the fish could successfully tolerate 50 percent sea water, one subgroup (IIj) of fry was exposed to 50 percent sea water 32 days after hatching. Based on the success of this early transfer, six subgroups (IIc through IIh) were placed in 50 percent sea water 36 days after hatching.

Mortality within the six subgroups (IIc through IIh) while in 50 percent sea water was low; in fact, it was lower than for the fresh-water fish (subgroups IIa and IIb).

Four of the six subgroups (IIc, IId, IIf, and IIg) were transferred to 75 percent sea water 54 days after hatching. The fish became wary and most did not feed until the second day after exposure. Mortality in 75 percent sea water was generally greater than mortality in 50 percent sea water within each subgroup (Table 4).

Four subgroups (IIc, IId, IIf, and IIg) were exposed to 100 percent sea water 66 days after hatching and once again exhibited behavior associated with osmoregulatory stress. Most fish would not feed for two days after exposure to full-strength sea water. The salmon were noticeably more wary in 100 percent sea water than they had been while in 75 percent sea water. Fish mortality, however, was lower in 100 percent sea water than in 75 percent sea water.

Subgroup IIh was transferred from 50 to 100 percent sea water 73 days after hatching. No mortality occurred, but the fish were under obvious stress. The changed behavior of this subgroup was more pronounced than that of any other subgroup transferred to 100 percent sea water. The salmon were extremely docile while in 50 percent seawater; but for 24 hours after exposure to 100 percent sea water, they would not feed. The fish also became extremely wary and

Table 4. Mortality of Group II salmon.

Age range during which the stated mortality occurred	Mortality in 50% sea water					Mortality in 75% sea water	Mortality during initial shock following exposure to 100% sea water		Mortality in 100% sea water after 47 hours without food[1]	Mortality in 100% sea water for the remainder of the experimental period	Total mortality
	18-30	32-46	36-117	36-73	36-54	54-66	66-68	73-75	87-89	76-86 90-117	
Subgroup											
IIa	—	—	—	—	—	—	—	—	—	—	5.3%
IIb	—	—	—	—	—	—	—	—	—	—	
IIc	—	—	—	—	0	1.6%	0	—	12.2%	13.8%	27.6%
IId	—	—	—	—	1.2%	1.2%	0.78%	—	12.8%	7.4%	23.38%
IIe	—	—	1.7%	—	—	—	—	—	—	—	1.7%
IIf	—	—	—	—	1.2%	3.5%	1.6%	—	13.3%	12.5%	32.1%
IIg	—	—	—	—	1.4%	2.8%	1.4%	—	12.26%	17.0%	34.86%
IIh	—	—	—	1.4%	—	—	—	0	4.25%	5.7%	11.35%
IIi	100%	—	—	—	—	—	—	—	—	—	100.0%
IIj	—	0	—	—	—	—	—	—	—	—	0

[1] All subgroups were starved for 47 hours due to endurance testing using a Category I test schedule; mortality occurred within 48 hours after feeding was resumed.

[2] Freshwater subgroups (IIa and IIb) were accidentally combined 66 days after hatching.

like all subgroups acclimated to 100 percent sea water, they remained wary to the end of the experimental period.

Mortality comparisons between the fresh water subgroups (IIa and IIb) and the sea water subgroups in Group II could not be made up to the end of the observation period. The fresh water fish succumbed to probable chlorine poisoning at 79 days of age due to temporary malfunction of the de-chlorination system.

For fish acclimated to 100 percent sea water, mortality was higher in Group II than in Group I. After the initial shock following exposure to 100 percent sea water, mortality for subgroups IIc, IId, IIf, IIg, and IIh in Group II was close to 11.1 percent, while mortality for subgroups Ic, Id, Ie, and If in Group I was about 1.7 percent.

Subgroups IIc, IId, IIf, and IIg which were acclimated to 100 percent sea water using the 50-75-100 percent acclimation route suffered a total mortality ranging from 23 to 35 percent (Table 4). However, subgroup IIh which was transferred directly from 50 to 100 percent sea water suffered an 11 percent total mortality.

Mortality for all Group II salmon is summarized in Table 4.

Disease was considered a possible factor contributing to the high mortality of Group II sea-water fish. Dead and dying salmon were examined microscopically but no evidence of disease was apparent.

Hyperosmotic Stress and Behavior

Fall chinook salmon 30 days of age[3/] and older exhibited little or no stress in 50 percent sea water, possibly because their body fluids were only slightly hypotonic to the external environment (Black, 1951). Increased hypertonicity of the environment was a probable cause of stress as indicated by changed behavior and increased mortality.

The behavior of my fish in 100 percent sea water just prior to death was similar to that described for juvenile steelhead trout by Conte and Wagner (1965). Steelhead which were unable to adapt to full-strength sea water developed a darkening of the skin, tended to tail,[4/] and demonstrated a series of gulping reflexes prior to death. Conte and Wagner examined some of the dying steelhead and found that their blood osmotic concentration and Na^+ and Cl^- ion concentrations were extremely high, indicative of the inability of the fish to osmoregulate. Mr. H. H. Wagner (personal communication) mentioned that dying chinook and coho salmon and steelhead in salt water demonstrated the same behavior. Dying chinook salmon in Group II demonstrated the skin darkening and tailing effect, while

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[3/] Age is expressed as days after hatching.

[4/] Refers to loss of body equilibrium.

most fish dying in Group I demonstrated the gulping reflex in addition to the darkening and tailing effect.

Hyperosmotic Stress and Survival

There is some evidence in the literature that small salmon exhibiting rapid growth can osmoregulate better than larger fish exhibiting slower growth (Wagner, Conte and Fessler, 1969). Parry (1961) found that better sea water survival and osmoregulatory ability occur in larger and older Atlantic salmon.

My Group II fish weighed more at the time of placement in 75 and 100 percent sea water than their Group I counterparts, due to faster growth resulting from slightly warmer water. For example, the 50-100 percent subgroup (IIh) from Group II averaged 1.567 grams while their Group I counterpart (subgroup Ic) averaged 1.154 grams when exposed to 100 percent sea water.

Group II fish were exposed to sea water at an earlier age than Group I fish. Subgroup IIh and subgroups IIc, IId, IIf, and IIg were exposed to 100 percent sea water seven and 14 days earlier, respectively, than their Group I counterparts.

It should be noted that total mortality for Group II sea water fish was 1.6 to 4.0 times higher than Group I mortality.

From the evidence examined, osmoregulatory ability was apparently a function of age in addition to growth and size in my experiments. Genetic differences may have played a part in determining the age at which salt-water tolerance occurred.

Juvenile steelhead and chinook salmon which cannot adapt to a hypertonic environment have been reported to die within ten days of initial exposure to sea water (Wagner, Conte and Fessler, 1969; Conte and Wagner, 1965; Weisbart, 1968). In both of my major groups of chinook salmon, but particularly for Group II, mortalities continued well beyond 30 days after initial exposure to full-strength sea water. This extended mortality may have resulted from a failure of osmoregulatory processes and a lack of tissue tolerance to the buildup of salts. Since the Group II fish experienced the higher mortality, they will be discussed in detail.

The Group II fish continued to exhibit symptoms of stress after initial exposure to full-strength sea water as indicated by their abnormal behavior, slow growth, and increased mortality. Tolerance of internal organs to a heavy salt load (Weisbart, 1968) may explain why some salmon which were under severe stress continued to survive for a month or longer. The fish which were under severe stress probably were forced to expend considerable energy to osmoregulate. Their incompletely developed osmoregulatory system and/or extrarenal secretory systems may ultimately have failed, allowing excess salts to enter the body tissues. This problem seemed to be particularly acute for fish held two days without food in 100 percent sea water.

During the two-day starvation period, most salmon were apparently using a fair amount of energy to osmoregulate. Upon being fed, the salmon consumed excess food over a brief interval of time (30 minutes). This may have caused energy needed for maintenance of osmoregulation to be used instead for digestion and assimilation. Osmoregulatory failure and death may have been the result. Such sudden energy transfer, though suicidal, may occur in young salmon, since they are not usually required at such an early age to cope with osmoregulation in full-strength sea water during an interval of rapid food consumption and assimilation.

GROWTH AND LENGTH-WEIGHT RELATIONSHIP

Growth and length-weight relationships were studied for chinook salmon held in fresh water and in 50 and 100 percent sea water. Similar observations were made on Group I and Group II fish.

Procedures

Fish were fed the Oregon Moist Pellet diet. Feeding frequency, pellet size, and amount of food fed per day were determined by following a feeding chart prepared by Bioproducts, Inc., Warrenton, Oregon. Both Group I and Group II fish were fed prior to yolk sac absorption in order to obtain good growth and food conversion (Palmer et al., 1951). Group I fish were first fed 31 days after hatching and Group II fish 33 days after hatching.

Salmon less than one gram in weight received mash five times daily. Salmon between one and two grams in weight received 1/32 inch pellets, while those over two grams received 1/16 inch pellets. Small fish were fed three to four times daily. Frequency of feeding was reduced to once or twice daily for larger fish.

For both major groups, the amount of food supplied was in excess of the amount the fish could consume. The amount of food presented at each feeding was calculated on the basis of a slight overfeed. The rearing tanks were cleaned every second or third day with a suction device to avoid the accumulation of food and fecal material, which can adversely affect salmon growth and stamina (Burrows, 1964).

During each feeding period, food was spread a little at a time fairly evenly over the water surface, so that all fish had a chance to satiate themselves. Salmon in tanks supplied with 100 percent sea water were easily frightened while being fed.

To minimize disturbance of the feeding fish, food was tossed into the tanks a little at a time from about three or four feet away.

Growth was determined twice monthly from measurements of length and weight. All subgroups were starved overnight immediately before the time of measurement.

Growth determinations were based on a sample of 30 fish from each subgroup. To obtain a sample, a large dip net was dragged through the schooled fish and a few fish at a time were transferred to a two-gallon dishpan until 30 fish had been isolated. These fish were then transferred a few at a time to a dishpan containing a gallon of anesthetizing solution (MS 222 and water).

Each fish taken from the anesthetizing solution was first measured for length in a "V"-shaped trough. Fish were then blotted on a damp paper towel to remove excess moisture before being placed in a 500 ml Erlenmeyer flask containing 100 grams of water. A triple-beam balance was used for weighing. After the anesthetized fish had been weighed, they were transferred to a dishpan for recovery and later returned to their rearing tank.

Data Analysis

A two-tailed Student's "t" test of the difference between mean values was employed to determine whether two subgroups of chinook salmon of the same age differed significantly in weight.

Growth rates were compared among the fresh-water and 50 and 100 percent sea-water subgroups in each major group by calculating values for instantaneous growth coefficients. Growth rate declined as weight increased (see subgroup Ib, Table 5); thus, the heavier fresh-water and 50 percent sea-water fish could not be compared to the smaller 100 percent sea-water fish of the same age. Growth coefficients were calculated and compared over weight ranges as similar as possible for the fresh-water and sea-water fish, regardless of age.

Growth rate was assumed constant within a given interval of time, and the following equation was used for calculating growth coefficients:

$$\text{Growth coefficient} = (\log W - \log W_o)/t \times 2.3026 \times 100$$

Table 5. Growth rates of Group I salmon.

Subgroup	Age (days)	Mean weight (grams)	Growth coefficient (rate): For 14 day interval	Growth coefficient (rate): Mean
Ia (0%)	92	1.66		2.26
	106	2.62	3.28[1/]	
	120	3.13	1.25	
	134	4.29	2.26	
Ib (0%-25%-50%)[2/]	92	1.61		2.09
	106	2.39	2.82	
	120	3.12	1.92	
	134	3.87	1.54	
Ic (0%-50%-100%)	92	1.50		1.34
	106	2.01	2.11	
	120	2.41	1.29	
	134	2.63	.63	
Id (0%-75%-100%)	106	1.68		.84
	120	1.92	.96	
	134	2.12	.71	
Ie (0%-50%-75%-100%)	106	1.74		1.20
	120	2.29	1.98	
	134	2.43	.43	
If (0%-25%-75%-100%)	106	1.80		.91
	120	2.16	1.31	
	134	2.33	.52	

1/ Growth rate of fresh-water fish (subgroup Ia) in the weight range 1.67 - 2.62 grams was contrasted with mean growth rates of fish in 100 percent sea water (subgroups Ic, Id, Ie, and If) having a similar weight range.

2/ Denotes the sequence of acclimation to sea water.

where W is the final weight of a subgroup; W_o is the initial weight of a subgroup; and t is the time interval in days; and 2.3026 computes $\log_e$.

Salmon weight as a function of length was also compared within a specific weight range among fresh-water and sea-water subgroups in each major group. The weight range corresponded to the range for subgroups which had been exposed to 100 percent sea water for at least two weeks before the end of the experimental period. Since fresh-water and 50 percent sea-water fish weighed more than 100 percent sea-water fish of the same age, weights of fresh-water and 50 percent sea-water fish determined at an earlier age were chosen to correspond to the weight range for 100 percent sea-water fish.

Regression lines were determined for weight as a function of length by using the following equation:

$$\log W = \log a + b \log L$$

where W is fish weight; L is fish length; a is a constant, the intercept of the weight-axis; and b is the regression coefficient (slope).

Regression coefficients were tested for significant differences among fresh-water fish and 50 and 100 percent sea-water fish. The following Student's "t" test for the difference between two regression coefficients was used:

$$t = (b_1 - b_2)/S\sqrt{1/\Sigma(X - \overline{X}_1)^2 + 1/\Sigma(X - \overline{X}_2)^2}$$

where $S^2 = (N_1 - 2)\, s_1^2 + (N_2 - 2)\, s_2^2/(N_1 + N_2 - 4)$;

b_1 and b_2 are regression coefficients for two different regression lines; N is the number of length-weight pairs for a given subgroup; s_1^2 and s_2^2 are the two subgroup variances for fish length; and X is fish length.

The table value of $t_{\alpha/2}$ was found at $N_1 + N_2 - 4$ degrees of freedom. This test is valid only if $s_1^2 = s_2^2$ as determined by an "F" test of the variances. Where $s_1^2 \neq s_2^2$, a slight variation of the "t" test was used and the degrees of freedom were determined by a simple formula (Bailey, 1959, pp. 99).

Due to the limitation in the computer program used, the maximum N for a regression line was 50. Certain sea-water subgroups contained more than 50 observations for length and weight. Length-weight pairs were discarded by the use of a table of random numbers until 50 remained. Only five or six length-weight pairs at the most were discarded in any single subgroup.

Observations on Growth

Growth of chinook salmon was definitely slowed by early exposure to sea water (Tables 5 and 6). Observations on mean weight vs. age (Figures 4 and 5) began at 50 days of age (Group I) and 51 days of age (Group II). At these approximate ages, certain subgroups were transferred to 50 percent sea water or a higher salinity.

Weight differences resulting from different growth coefficients (rates) were compared among fish of Group I held in fresh water and in 50 and 100 percent sea water. All subgroups had approximately the same mean weight at 50 days of age.

Subgroup Id had an extremely slow growth rate in 100 percent sea water (Table 5). Even after 30 days in 75 percent sea water, this subgroup weighed significantly less ($p < 0.001$) than the fresh-water subgroup (Ia).

Subgroup If grew at a rate which was almost one-fourth that of the fresh-water subgroup. This subgroup also weighed significantly less ($p < 0.002$) than the fresh-water fish after only 14 days in 75 percent sea water.

Subgroup Ie grew at a rate which was one-half that of the fresh-water subgroup. This subgroup weighed significantly less than the fresh-water subgroup after exposure to 100 percent sea water ($p < 0.001$).

The highest mean growth rate in 100 percent sea water was recorded by subgroup Ic. It was still only one-half that of the fresh-

Table 6. Growth rates of Group II salmon.[1/]

Subgroup	Age (days)	Mean weight (grams)	Growth coef. (rate) For 14 day interval	Growth coef. (rate) Mean
IIa and IIb (0%)	65 79	1.02 1.62	3.15	——
IIc (0%-12.5%-50%-75%-100%)	65 79 93	1.15 1.54 1.74	2.06 1.16	1.61
IId (0%-12.5%-50%-75%-100%)	65 79 93	1.20 1.59 1.76	2.04 .69	1.37
IIe (0%-12.5%-50%)	65 79 93	1.32 1.89 2.47	2.51 1.91	2.21
IIf (0%-25%-50%-75%-100%)	65 79 93	1.15 1.44 1.73	1.61 1.32	1.47
IIg (0%-25%-50%-75%-100%)	65 79 93	1.17 1.49 1.81	1.74 1.12	1.43
IIh (0%-25%-50%-100%)	79 93	1.79 2.01	.84	——

[1/] Within similar weight ranges, mean growth rates of fish (subgroups IIc, IId, IIf, IIg and IIh) in 100 percent sea water were contrasted with growth rates of fresh-water fish (subgroups IIa and IIb) and fish in 50 percent sea water (subgroup IIe).

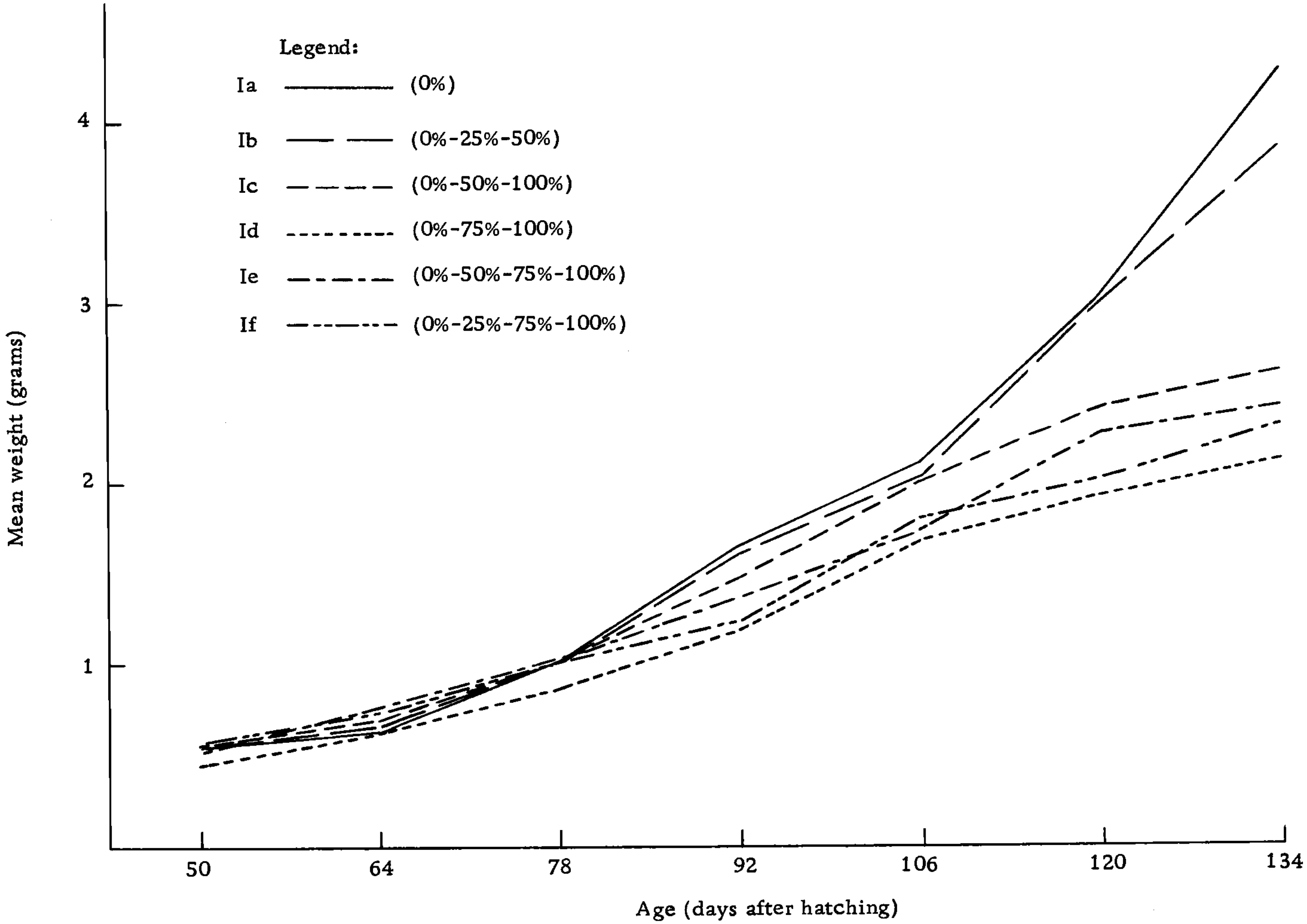

Figure 4. Growth of Group I salmon.

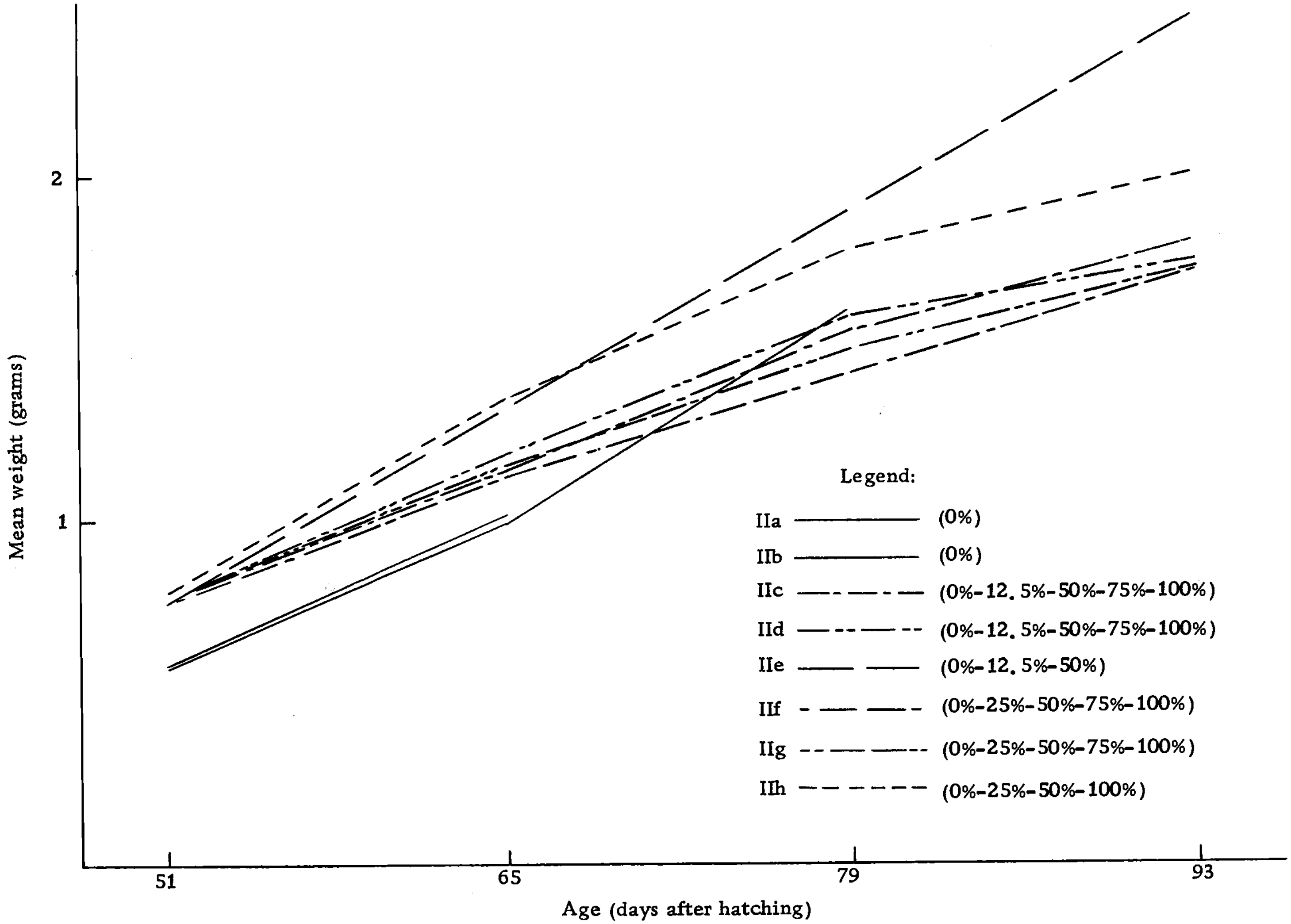

Figure 5. Growth of Group II salmon.

water subgroup. Subgroup Ic weighed less ($0.05 > p > 0.01$) than fresh-water fish after 14 days in 100 percent sea water.

The 50 percent sea-water subgroup (Ib) grew almost as fast (Table 5) as the fresh-water subgroup (Ia). However, fish in 50 percent sea water weighed less ($p < 0.001$) than fresh-water fish at 134 days of age.

The differences in mean weight between the fresh-water subgroups and all 100 percent sea-water subgroups were quite apparent (Figure 4) at the end of the experimental period (134 days).

At the time of the first growth measurement (51 days of age) of Group II fish, both fresh-water subgroups (IIa and IIb) weighed significantly less ($p < 0.001$) than the other six subgroups (IIc through IIh) which were all in 50 percent sea water at the time.

At 65 days of age, after four of the six subgroups (IIc, IId, IIf, and IIg) were exposed to 75 percent sea water for 14 days, the fresh-water subgroups still weighed less ($0.05 > p > 0.01$) than any sea water subgroup. However, both subgroups (IIe and IIh) remaining in 50 percent sea water weighed significantly more ($0.01 > p > 0.005$) than the subgroups in 75 percent sea water.

One day later (66 days of age), the rearing tank partition separating the two fresh-water subgroups collapsed, combining both subgroups. Both subgroups were practically identical in weight so no harm was done. This explains the single line in Figure 5 for fresh-water fish (subgroups IIa and IIb) after 65 days of age.

At 79 days of age, the four subgroups (IIc, IId, IIf, and IIg) had been exposed to 100 percent sea water for 14 days and subgroup IIh had been in 100 percent sea water for seven days. At this time, the fresh-water fish weighed more than subgroups IIc, IId, IIf, and IIg. Subgroups IIh and IIe weighed more ($0.05 > p > 0.001$) than the fresh-water fish.

After 21 days in 100 percent sea water, subgroup IIh weighed less ($0.01 > p > 0.005$) than subgroup IIe. Subgroup IIh weighed more ($p < 0.01$) than any other subgroup (IIc, IId, IIf, and IIg) in 100 percent sea water.

Observations on Length-Weight Relationships

In Group I, length-weight relationships were compared between fresh-water fish (subgroup Ia) and three subgroups (Ic, Id, and Ie) acclimated to 100 percent sea water (Figure 6) within a weight range of 2.0 - 3.0 grams.

Subgroups Ic and Id weighed a little more than subgroup Ia at a given length. The regression coefficient for subgroup Id was not significantly larger ($p > 0.01$) than that of subgroup Ia. The coefficient for subgroup Ic was not significantly smaller ($p > 0.05$) than that of subgroup Ia; therefore, as fish length increased, the change in weight for a given change in length was slightly smaller for subgroup Ic than for subgroup Ia.

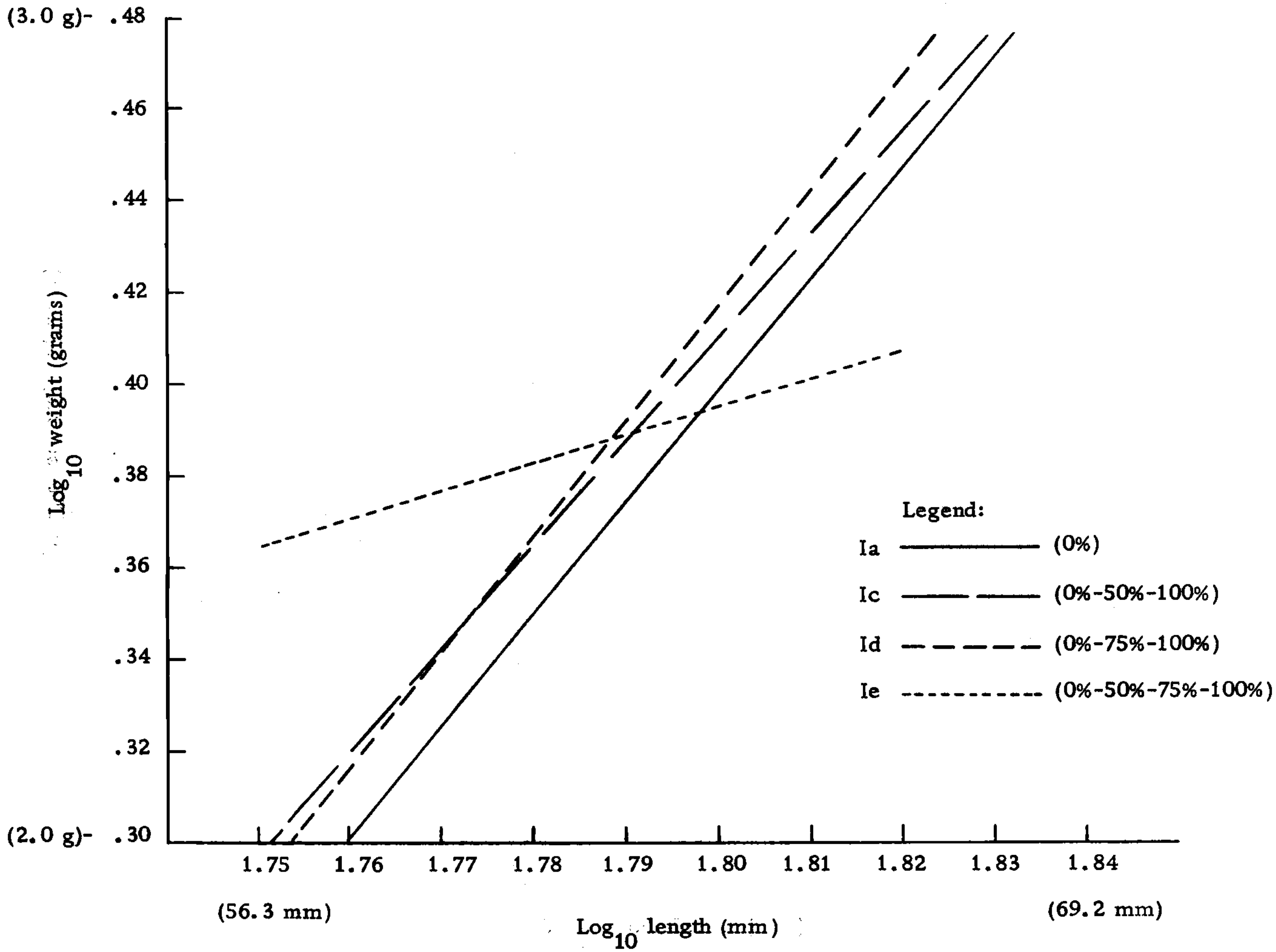

Figure 6. Length-weight relationship of Group I salmon.

The regression coefficient for subgroup Ie was significantly less ($p < 0.01$) than that for subgroup Ia. However, fish in subgroup Ie less than log 61.95 mm. long (log = 1.79, Figure 6) weighed more at a given length than fish in subgroup Ia, but those over 61.95 mm. weighed less at a given length.

In Group II, length-weight relationships were compared between 50 percent sea-water fish (subgroup IIe) and three subgroups (IIf, IIg, and IIh) acclimated to 100 percent sea water. Subgroups IIf and IIg were examined in the 1.5-2.0 gram weight range and subgroup IIh was examined in the 1.5-2.5 gram weight range.

Fish weights at a given length in subgroups IIf and IIg were slightly higher than weights in subgroup IIe (Figure 7). Regression coefficients for subgroups IIf and IIg were smaller (significantly for IIg at $p < 0.01$) in comparison to the coefficient for subgroup IIe.

Fish in subgroup IIh weighed slightly more at a given length than fish in subgroup IIe (Figure 8). The regression coefficients for both subgroups were almost identical ($p > 0.05$).

Effect of Salinity on Growth

In Group II, the fresh-water salmon weighed significantly less ($p < 0.01$) than the salmon exposed to dilute sea water (12.5 and 25 percent) at the time of the first growth observation (51 days after hatching) (Figure 5). All Group II salmon were from the same initial

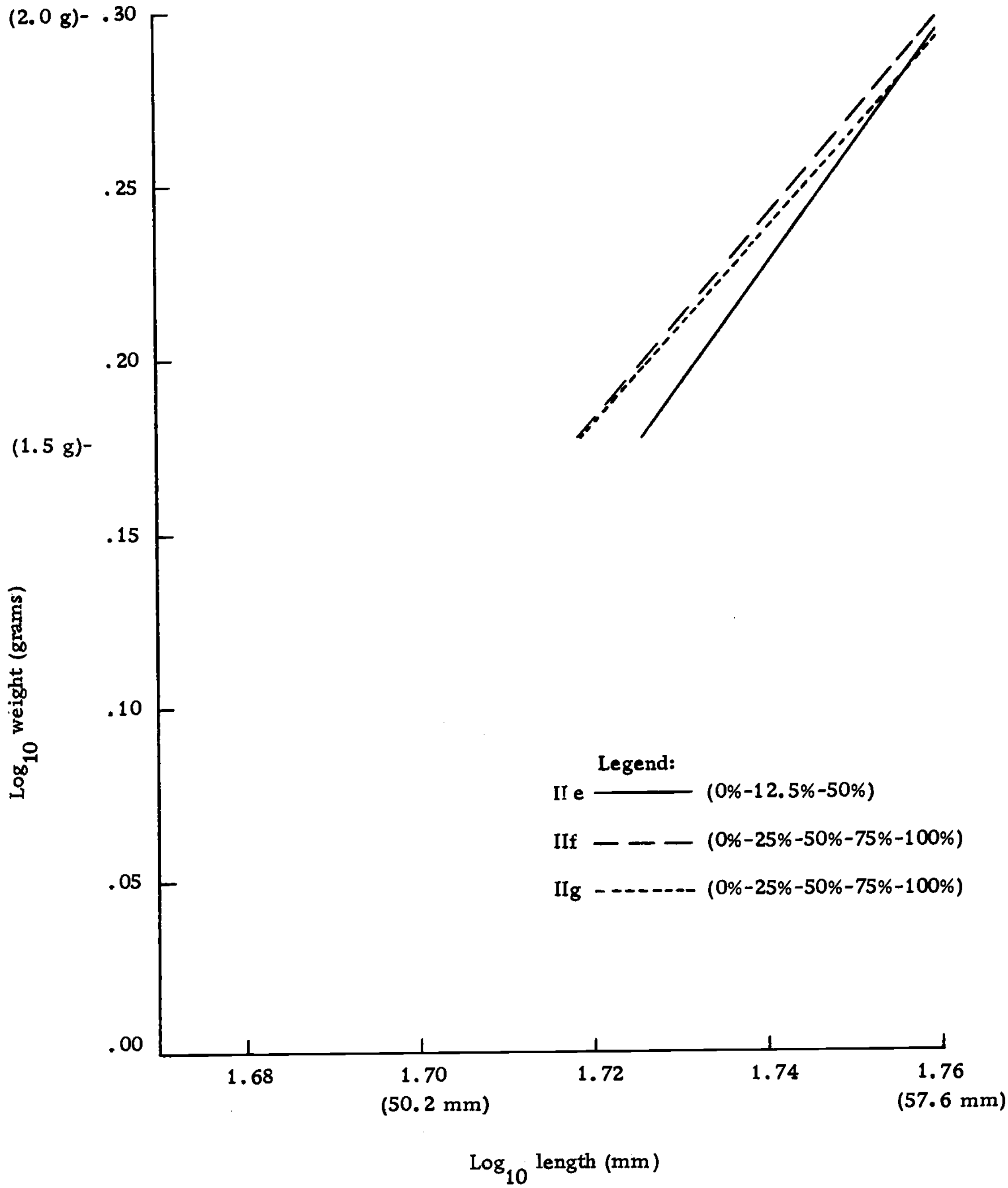

Figure 7. Length-weight relationship of Group II salmon.

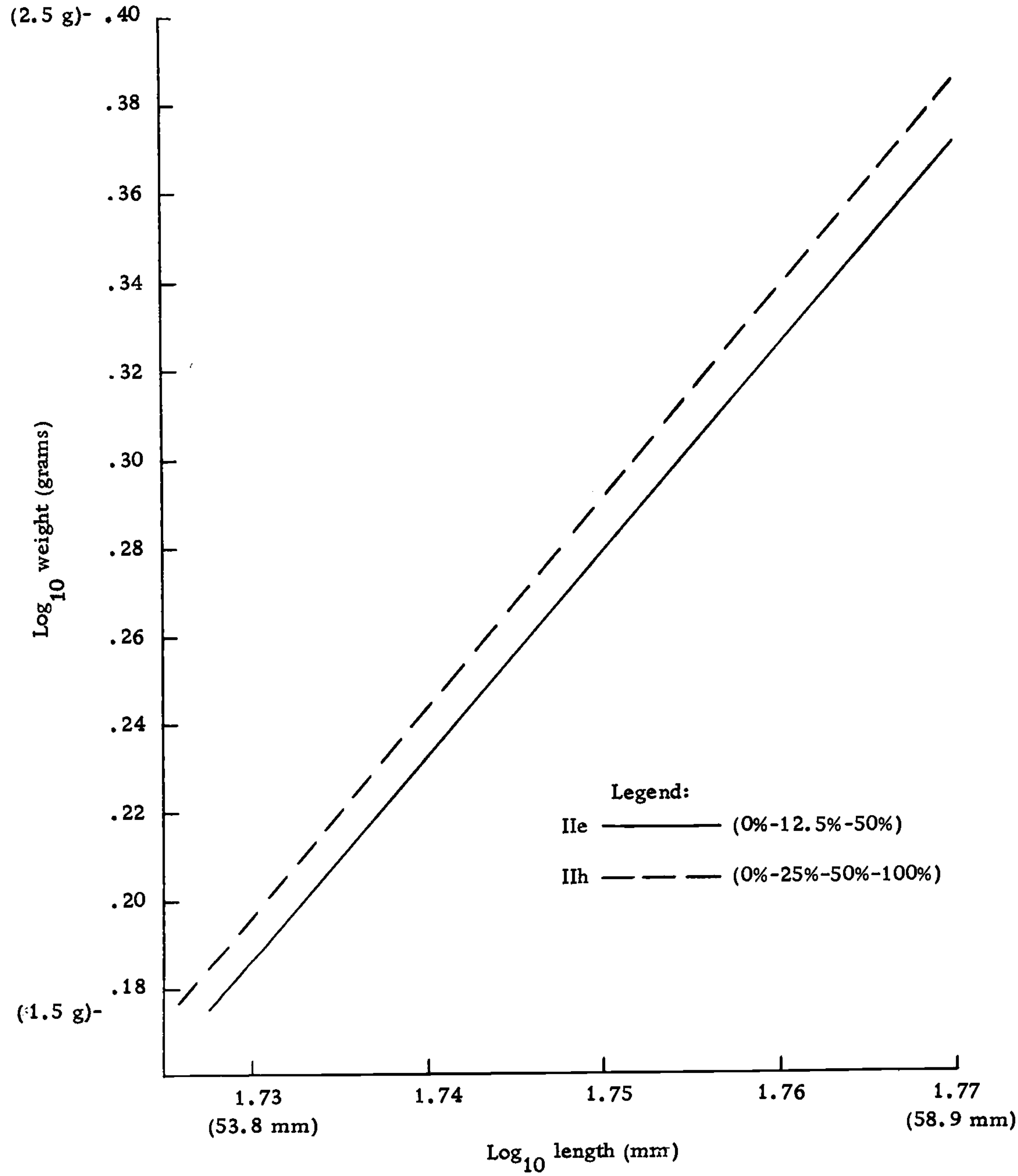

Figure 8. Length-weight relationship of Group II salmon.

fresh water population and were subjected to identical treatment insofar as is known. All fish in the egg and alevin stage were shielded from natural and fluorescent light to prevent any effect of light on salmon development as shown by Eisler (1961).

The size difference at 51 days of age can be explained by the difference in initial feeding times. Salmon in 12.5 and 25 percent sea water were more active and began to feed at an earlier age than salmon in fresh water. Although the fish were sampled selectively for the rearing pans due to the presence of many anomalies in the original population, and probably the most active fish escaped from the rearing pans, the fact that early exposure to dilute sea water may enhance growth and development should not be overlooked.

In an isotonic environment, fish would expend little energy in maintaining salt and water balance. Since energy required for osmoregulation would be nil, more food energy could be used for growth than in a fresh-water or ocean environment. Canagaratnam (1959) has shown that small coho salmon (0.6-1.1 grams) have a higher growth rate in salt water of 12‰ and 18‰ than in fresh water. Bullivant (1961) showed that one-year-old chinook salmon grew faster in 50 percent sea water (17.7‰) than in fresh water. Apparently, 50 percent sea water is approximately isotonic to salmon body fluid, at least for chloride ions as shown for chum and pink salmon, and apparently for coho salmon by Black (1951).

In my experiments, chinook salmon held in 50 percent sea water in both major groups had lower growth rates than fresh-water fish. Salinity and possibly water temperature explain the lack of a high growth rate for the former salmon. Due to mechanical problems and natural fluctuations in sea-water salinity, the salinity of 50 percent sea water was not constant (Tables 7 and 8) during the experimental period. Also, for about 30 days the temperature of 50 percent sea water was lower by about 1.0°C than the fresh-water temperature.

In my experiments, all salmon acclimated to 100 percent sea water grew at a slower rate than fish in fresh water or in 50 percent sea water. Consequently, they were significantly smaller than fish in fresh water and in 50 percent sea water prior to completion of the growth experiments. This agrees with the findings of Bullivant (1961) who has shown that one-year-old chinook salmon in sea water (35.4‰) had a maximum growth rate half that of a comparable group of fresh-water salmon.

The low growth rate of salmon in 100 percent sea water is most likely a result of osmoregulatory stress. Within 24 hours after salmon were exposed to 100 percent sea water, there was an apparent change in their food consumption. The salmon would not eat for one or two days after exposure and were easily frightened. After this short period the salmon began to feed but did not consume as much

Table 7. Salinity and temperature during growth and stamina[1] experiments with Group I salmon.

Salinity				
Desired salinity (‰)	Growth period (‰)		Endurance test period (‰)	
	Mean	Range	Mean	Range
8 (25 percent)	8.9	7.6-9.5		
16 (50 percent)	17.3	14.9-21.7	16.8	13.1-19.7
24 (75 percent)	23.9	21.0-26.0		
32 (100 percent)	32.9	30.2-36.2	32.2	30.3-33.3

Temperature (°C)						
	Growth period				Difference between fresh water and sea water temperatures	
	Fresh water		Sea water			
Dates	Mean	Range	Mean	Range	Mean	Range
February 27 - April 28	10.7	8.0-13.0	10.8	8.0-13.0	0.4	0-2.0
April 28 - May 22	13.5	11.5-16.0	11.8	9.5-12.0	2.5	0.5-4.0

[1] Water temperatures during stamina tests are shown in Table 11.

Table 8. Salinity and temperature during growth and stamina experiments with Group II salmon.

Salinity				
Desired salinity (‰)	Growth period (‰)		Endurance test period (‰)	
	Mean	Range	Mean	Range
16 (50 percent)	18.4	15.6-23.4	16.7	13.0-20.2
24 (75 percent)	24.6	20.5-26.0		
32 (100 percent)	32.5	30.2-34.9	32.2	30.3-33.3

Temperature (°C)						
	Growth period				Difference between fresh water and sea water temperatures	
	Fresh water		Sea water			
Dates	Mean	Range	Mean	Range	Mean	Range
April 8 - April 28	11.9	11.0-13.0	12.0	9.0-13.0	0.3	0-1.0
May 1 - May 20	13.4	11.5-16.0	10.9	9.5-12.0	2.5	0.5-4.0

food relative to weight as the fish in fresh water and in 50 percent sea water.

My salmon were exposed to 100 percent sea water at an earlier age than they would normally be in nature and probably required more energy to attain and maintain osmoregulatory "homeostasis" than salmon totally adapted to sea water. If any excess energy had to be taken from the energy pool needed for osmoregulation, such as for the digestion and assimilation of excess food, the health of the fish could be in jeopardy. This was already discussed in relation to salmon mortality in 100 percent sea water resulting after the fish consumed a large amount of food after their starvation period for stamina testing. Decline in food consumption may be a protective mechanism for the salmon not fully adapted to 100 percent sea water, as long as the fish are not subjected to an added stress situation, like starvation.

Food consumption and wary behavior appear to be a function of osmoregulatory stress. In the laboratory, fresh-water salmon became increasingly wary as their size increased. Fresh-water alevins and fry were friendly to a fault, but after absorbing their yolk they became increasingly wary and readily sought shelter when an observer approached.

Wary behavior seems to be a function of hunger in addition to size. In my experiments, unfed salmon regardless of size exhibited less fear or wariness to an observer than fish which had recently been fed. Even when hungry, however, the young salmon in 100 percent sea water appeared wary compared to hungry salmon in fresh water. The salmon held in sea water seemed to be hungry in the morning, expressed relatively little fear, and fed as well as any of the fresh-water fish of similar size. Unlike the fresh-water fish, however, the sea-water fish apparently did not feed as actively during the rest of the day and tended to remain quite wary.

The food digestion rate for my salmon in 100 percent sea water may have been slowed since much energy was needed for osmoregulation. Hence, the fish may have been essentially "full" all day from one meal and exhibited the expected wary behavior.

It was not apparent whether the slow growth of salmon in 100 percent sea water resulted from their rejection of the moist pellet diet. But salmon in 100 percent sea water fed live amphipods had a lower growth rate than did fresh-water salmon (Bullivant, 1961).

In general, the length-weight relationship of fish in 100 percent sea water was similar to that of fish in fresh water and in 50 percent sea water and was apparently not affected by osmoregulatory stress.

ENDURANCE

In addition to the observations on behavior, survival, and growth, stamina of juvenile chinook salmon acclimated to 100 percent sea water was compared to that of fish from fresh or brackish water.

Methods and Materials

Swimming Tunnels

Endurance was measured by forcing salmon to swim against a series of water currents in two swimming tunnels.

The endurance test apparatus (Figures 9 and 10) contains two six-foot-long plexiglas swimming tunnels, each having a diameter of 2.5 inches. Each tunnel contains three 0.25 inch mesh screens in the forward section and an electrical fish barrier with a rotating screen in the rear section. A 2.0 inch I.D. PVC valve in front of each tunnel controls water flow.

Water is delivered to the tunnels by a centrifugal pump powered by a 0.5 HP motor and having a 2.0 inch I.D. intake and discharge. A maximum water velocity of about 1.62 ft/sec (30 gpm) was obtained in each tunnel.

A 30 psi water pressure gauge was placed in front of each valve used to control water flow in the tunnels. The gauges in conjunction

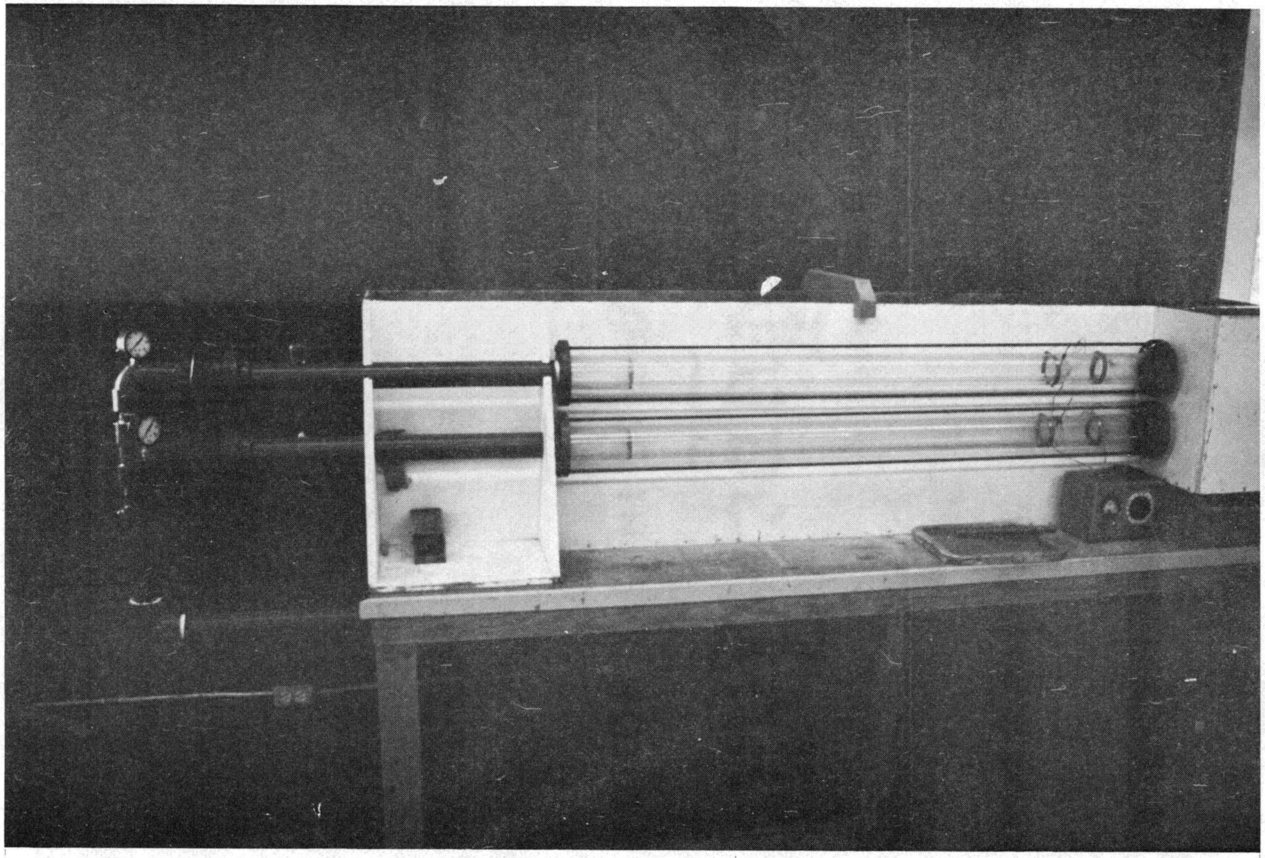

Figure 9. Apparatus used for endurance testing of juvenile salmon.

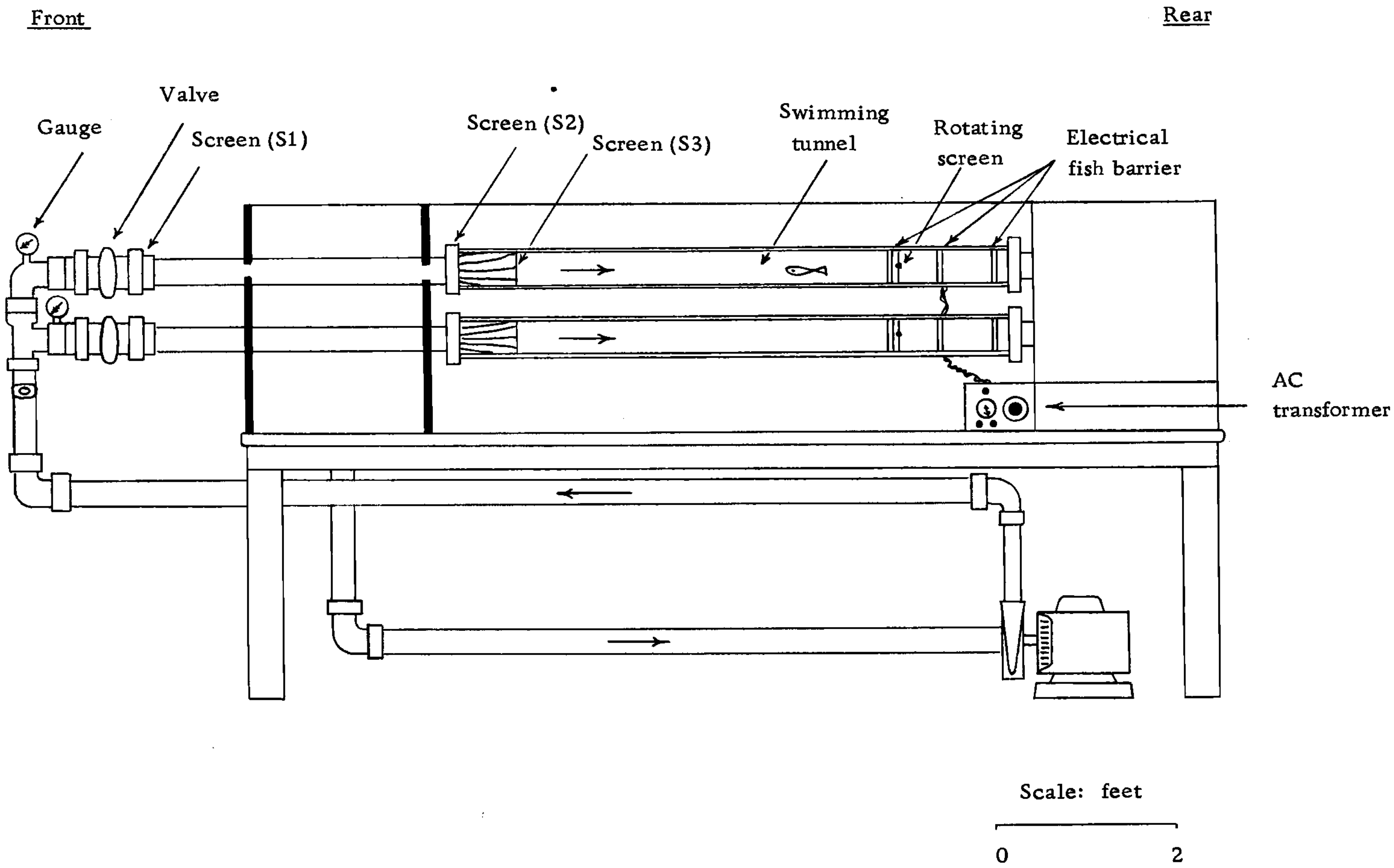

Figure 10. Diagram of the apparatus used for stamina testing of juvenile salmon.

with an indicator board[5] located under the handle of each valve aided in determining water velocities for each endurance test.

The endurance apparatus can recycle water or function with only partial recycling. When complete recycling was used, water temperature rose 1.0 C per hour of running time. With partial recycling, it was possible to keep the water temperature within 0.5 C or less of the initial temperature after several hours of operation.

A plastic-coated 0.25 inch mesh screen (S1)[6] was situated about 1.0 inch in back of each 2.0 inch I.D. PVC ball valve to minimize turbulence (Figure 10). An identical screen (S2) at the junction of the 2.0 inch I.D. PVC pipe and the forward section of a swimming tunnel served to diminish turbulence and to prevent salmon from entering the valve. Another screen (S3) is located about 10.0 inches in back of screen S2. Kerr (1953) noted that chinook salmon were able to detect areas of low water velocity (eddys). Screen S3 was installed to prevent the fish from seeking a small eddy in back of S2.

Salmon were prevented from leaving the rear of a swimming tunnel by a moveable rotating screen and an AC electric field. The

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[5] A plywood board containing markings which line up with a mark on each valve handle to indicate a particular water velocity in each swimming tunnel.

[6] (S1) = screen # 1.

electric field[7/] was confined to an area within three 0.5 inch wide stainless steel bands located in the rear section of each tunnel (Figure 10). The middle band acted as an opposite pole for the two outside bands. Desired voltages were maintained by a 0-150 volt AC transformer. During actual test runs, a maximum of 10 volts was supplied to the bands.

During preliminary trial endurance tests, the electric field failed to adequately perform its function of forcing the fish to swim against the current until exhausted. After placing salmon in the tunnel, a 10.0 volt electric field was first obtained in standing water. Fish usually were restless and some appeared to be panicky. Some frightened fish darted into the electric field, were badly shocked, and of no further use in the test.

Lower voltages which mildly shocked the fish were tried, but this failed to solve the problem. When a 5.0 volt electric field was used, frightened fish either swam out of the tunnel through the field or remained in the field long enough to become partially stunned and of no further use. Some of the remaining fish detected the 5.0 or 10.0 volt electric field, turned around, and remained in the tunnel.

Following an initial period of adjustment to the swimming tunnel,

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[7/] The electric field is similar to that used in the swimming tunnel at the U.S. Bureau of Sport Fisheries and Wildlife Salmon-Cultural Laboratory at Longview, Washington.

a low velocity water flow was introduced and the test fish were given an opportunity to contact the electric field. Most salmon eased back tail first to the rear section of a tunnel, felt the shock, and swam away from the electric field. However, as many as 50 percent of the salmon placed in a tunnel passed through the electric field prior to exhaustion and were removed from the test.

To reduce the number of fish passing through the electric field prematurely, a rotating 0.25 inch mesh, plastic-coated screen was installed in each tunnel about 0.125 inch behind the point at which the fish first came in contact with the electric current. The rotating screen consists of a round plastic bar which is threaded through a loose circular screen inside each swimming tunnel. The bar fits between two small plastic taps that are externally screwed into each tunnel and can be rotated perpendicular to the axis of the tunnel by an attached handle.

The rotating screen allowed a fish to turn around once it contacted the electric field. Hence, the problem with frightened fish leaving the tunnel was solved. After the test fish had been placed in a tunnel, the rotating screen was closed. It was opened only to remove exhausted fish during a test.

Two swimming tunnels were used in simultaneous tests of two samples of fish from the same subgroup to permit replicate observations.

Test Schedules

Three test schedules[8/] were utilized to find the most useful technique for testing endurance. Two test schedules of identical duration but employing different water velocities were used in preliminary comparisons with small and large fish. The older and larger fish from Group I were tested against higher water velocities than Group II fish (Table 9). Test schedules for both Group I and Group II salmon involved a maximum duration of 3.5 hours under these "Category I" test schedules.

Category I test schedules were entirely experimental and were found, unfortunately, to have disadvantages. Each schedule was specific for salmon in a certain major group and made invalid any direct comparison of the endurance of Group I vs. Group II salmon. Performance comparisons of subgroups within a major group were difficult, because the intervals between water velocities and the duration of each velocity were not equal in each test schedule. Due to longer time periods at higher water velocities in each Category I test schedule, large salmon were permitted longer swimming times than they would have had in a schedule allowing an equal duration for each water velocity.

[8/] A test schedule is a table giving a breakdown of water velocities and their respective time intervals.

Table 9. Endurance test schedules.

Category I

Water velocity (ft/sec)	Time interval (min) at each velocity
Test schedule for Group I salmon	
0.65	10
0.81	10
0.86	10
1.08	120
1.18	60
1.24	remainder of test time
Test schedule for Group II salmon	
0.65	5
0.81	175
0.86	30
1.08	remainder of test time

Category II

Test schedule for Group I and Group II salmon

Water velocity (ft/sec)	Time interval (min) at each velocity
0.54	5
0.81	15
1.08	15
1.35	15
1.62	15

Because of problems with Category I test schedules, a "Category II" test schedule[9/] (Table 9) was adopted for further tests with Group I and Group II salmon. This schedule made use of identical time periods for each water velocity above 0.54 ft/sec. Increments in velocity were held at 0.27 ft/sec between adjacent time periods. With the Category II schedule, it was possible to complete tests with all subgroups of a major group in one day; whereas, two days were required previously with Category I schedules.

Performance comparisons of the different subgroups within each major group were facilitated with the Category II schedule and, of equal importance, comparisons of Group I to Group II salmon were possible.

Selection and Placement of Fish

The sequence in which subgroups were selected for testing was determined by choosing a card at random from a card deck containing subgroup tank numbers. The first card chosen indicated the subgroup to be tested first. After a tank was chosen, its card was excluded from the deck.

[9/] The Category II test schedule is a modification of the stamina test procedure used at the U. S. Bureau of Sport Fisheries and Wildlife Salmon-Cultural Laboratory at Longview, Washington.

The tendency for all fish in a tank to school when disturbed was used to advantage when selecting fish for testing. A large dip net was used to collect most of the schooled salmon, and while the net was half submerged, a small dip net transferred an equal number of fish into separate small containers. One container was labeled "top" and the other "bottom" to correspond to the positions of the two swimming tunnels. When sampling a tank, the placement of the first of two fish samples into either the "top" or "bottom" container was determined by flipping a coin.

Under Category I test schedules, 10 fish from each subgroup in Group I were placed in each swimming tunnel. In Group II, only eight fish were placed in each tunnel for a test run. Under the Category II schedule, 12 fish from the subgroup tested were placed in each tunnel. For all tests, salmon were placed in the bottom swimming tunnel first. The following procedure was used for placing fish in the tunnels. One end of a 0.75 inch I.D. tygon tube was inserted into the rear of a tunnel past the series of electric bands, and a plastic funnel was then inserted into the other end of the tubing (Figure 11). Fish were next placed into the tygon tube by dip netting them into the funnel. Most salmon from each sample entered the tubing head first, but this preferred position could not be obtained for all fish due to the excessive amount of time required just to get the fish into the swimming tunnel. When all fish were in the tygon tubing, the

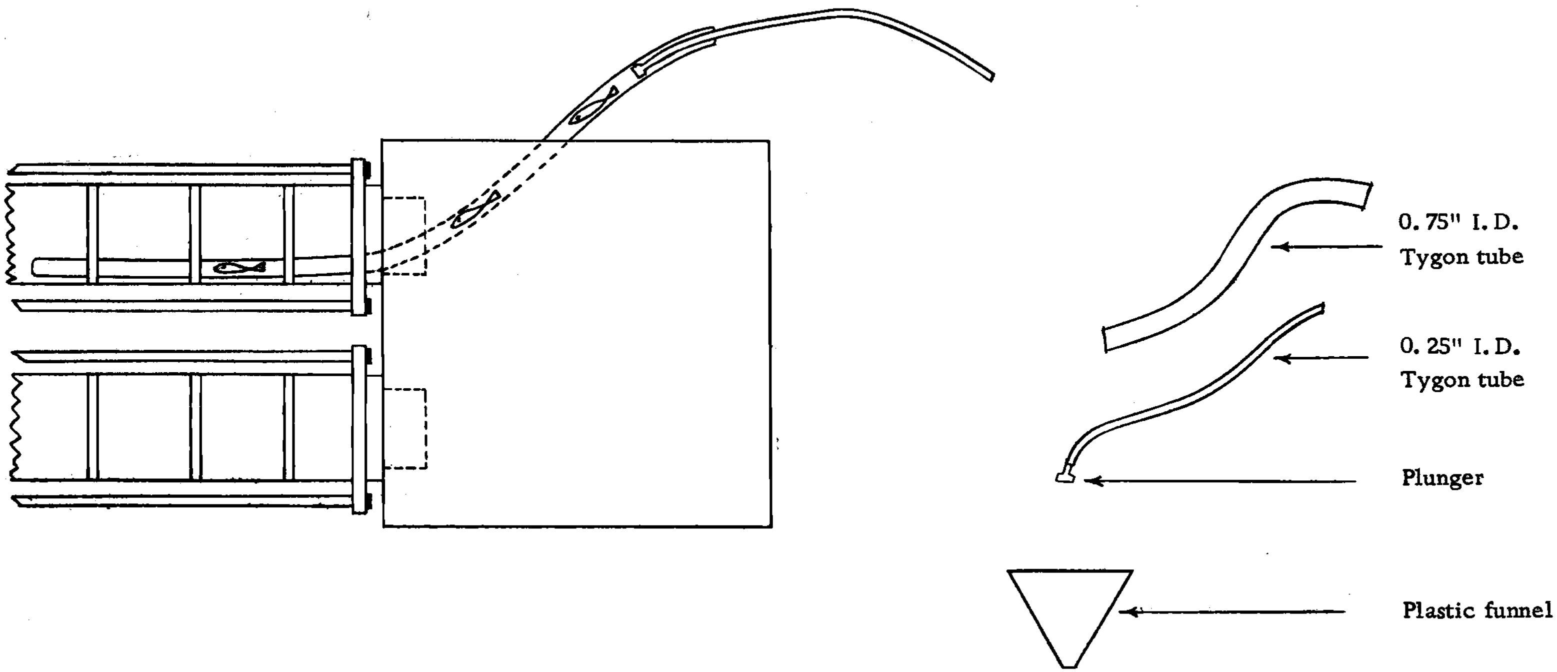

Figure 11. Placement of fish into a swimming tunnel.

funnel was removed, and a plunger was inserted to gently push the fish into the tunnel. Usually a small number of fish swam out of the tunnel before the rotating screen could be closed. These fish were collected and replaced into the tunnel. The total amount of time required to place all fish into both tunnels ranged from four to 20 minutes with a mean of 10 minutes.

Endurance Test Procedure

The temperature of fresh water usually remained within 2.0 C of the sea-water temperature during the experimental period.

All salmon in a major group were not fed for 24 hours prior to testing to insure that the energy needed for food digestion and assimilation could not affect fish performance (Thomas, Burrows and Chenoweth, 1964).

Immediately following their placement in the swimming tunnels, fish were allowed to rest. The length of the rest period varied according to the endurance schedule employed. Under Category I endurance schedules, a 10 minute rest period for Group I salmon and a six minute rest period for Group II salmon were employed. All fish tested under the Category II endurance schedule received a 15 minute rest period. Since the salmon were placed in the bottom tunnel first, they had the longest rest period before they were forced to exercise.

Bams (1967) noted that an adaptation period prior to swimming performance tests was effective in teaching fish to avoid a downstream electric grid and retaining screen. Hence, during my tests, a 5.0 volt electric field was introduced across the bands for the duration of the rest period. Any salmon swimming near the rotating screen during this period quickly learned to avoid it during the actual test.

After the rest period, the pump was turned on and off intermittently at the lowest water velocity until all fish were swimming against the water current. The pump was then allowed to run continuously to initiate the test. It was assumed that most, if not all, fish had encountered the electric field by this time, and the voltage was increased from 5.0 to 10.0 volts to provide an extra incentive for the salmon to swim until exhausted.

During each test run, the time was recorded when individual fish could no longer swim against the water current. Each exhausted fish was removed and transferred to a container filled with standing water. At the completion of a test, all fish were weighed and measured for length in order to relate fish size with stamina. The salmon were not returned to their rearing tank to avoid their possible re-selection for future stamina tests (Brett, Hollands and Alderdice, 1958; Thomas, Burrows and Chenoweth, 1964).

Performance Comparisons

In the endurance experiments, a performance comparison was intended of the fish held in fresh water to those in 50 percent and 100 percent sea water in the same major group. In order to make comparisons, a performance index used by Thomas et al. (1964) was adopted. This index is half the summation of the time required for 25 and 75 percent of the fish to leave the swimming tunnel. Therefore, the index measures the swimming performance of only the middle 50 percent of the fish and thus excludes the strongest and weakest fish.

Observations on Endurance

Because the two categories of test schedules used in these experiments differed considerably, the results will be discussed separately.

Results with Category I Test Schedules

Due to the disadvantages in the Category I test schedules, analysis of the data was difficult and no final conclusions could be drawn. Data on performance were analyzed by comparison of wt./p.i.[10/] ratios within each major group. Since salmon endurance

[10/] Wt./p.i. = weight/performance index.

relative to weight decreases as fish weight increases (Brett, 1965), fish from the same population would exhibit increased wt./p.i. ratios as they increase in weight.

In Groups I and II, wt./p.i. ratios for five sea-water subgroups increased as fish weight decreased (Table 10). The wt./p.i. ratios obtained with Group I fish were considerably larger than those for Group II fish, probably resulting from different water velocities used in the Category I test schedules (see Table 9).

Results with the Category II Test Schedule

The Category II test schedule allowed a more meaningful analysis of test results. Performance data were analyzed for the subgroups tested in each major group and for the Group I and Group II subgroups.

Linear regressions of performance index on fish weight (Figures 12 and 13) were compared between the Group I and Group II subgroups tested. Thomas *et al*. (1964) have found a straight line relationship between fish weight (log) and performance index (Figure 14).

Performance comparisons of salmon in 100 percent sea water to salmon in fresh water and in 50 percent sea water was difficult because of the larger size of the later fish at the time of testing and the loss of all fresh-water fish in Group II prior to testing.

All salmon in 100 percent sea water demonstrated lower

Table 10. Endurance test results for Group I and Group II salmon using the Category I test schedules.

Subgroup	Salinity[1]	Date tested	Performance index[2] (minutes)	Mean weight (grams)	Wt./p.i. ratio	Swimming tunnel water temp. (°C) start	finish
			GROUP I				
Ia	0	May 16	97.1	3.59	.036	13.0	15.0
Ib	50	May 17	61.3	3.46	.056	13.25	15.0
Ic	100	May 16	23.1	2.74	.100	12.0	12.5
Ie	100	May 16	17.1	2.37	.138	12.25	13.0
			GROUP II				
IIe	50	May 13	203.1	2.01	.010	12.5	13.0
IIh	100	May 14	121.3	1.78	.015	12.0	12.5
IIc	100	May 13	38.3	1.66	.044	11.0	11.5
IIf	100	May 14	48.9	1.55	.032	11.25	11.75

[1] Denotes the percent salinity of the water in which the fish lived and were tested.

[2] Mean performance index for both swimming tunnels.

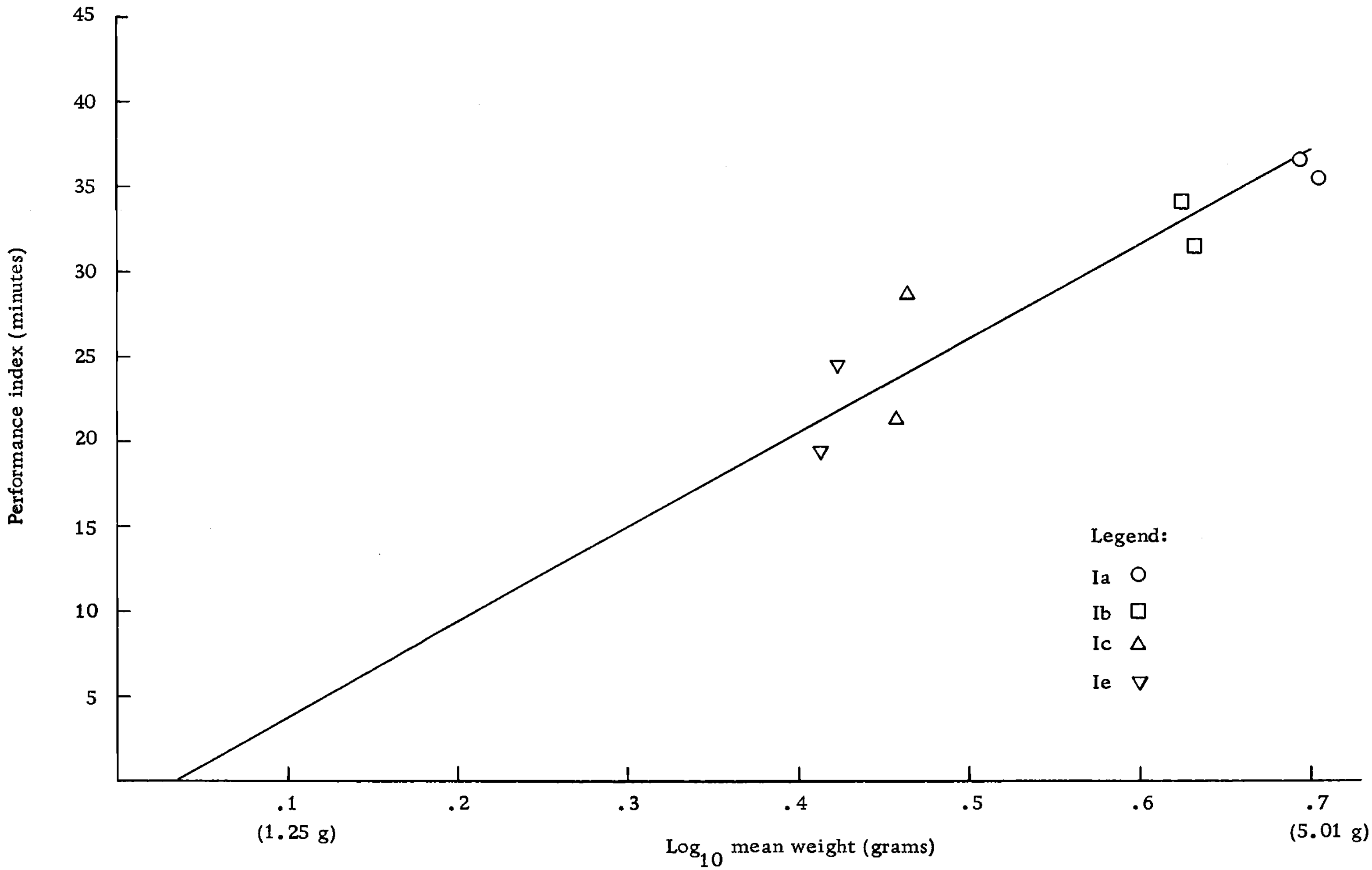

Figure 12. Endurance of Group I salmon relative to weight.

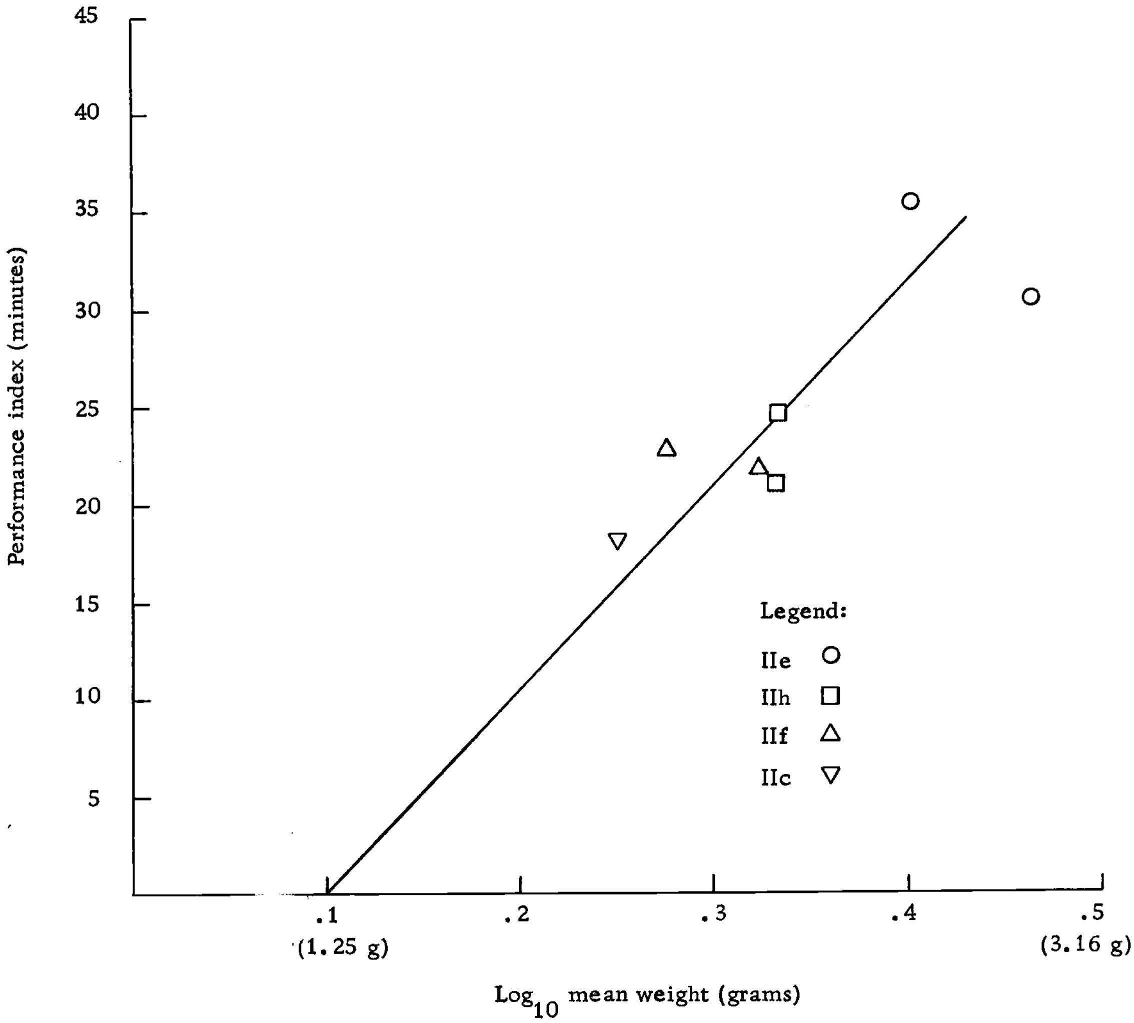

Figure 13. Endurance of Group II salmon relative to weight.

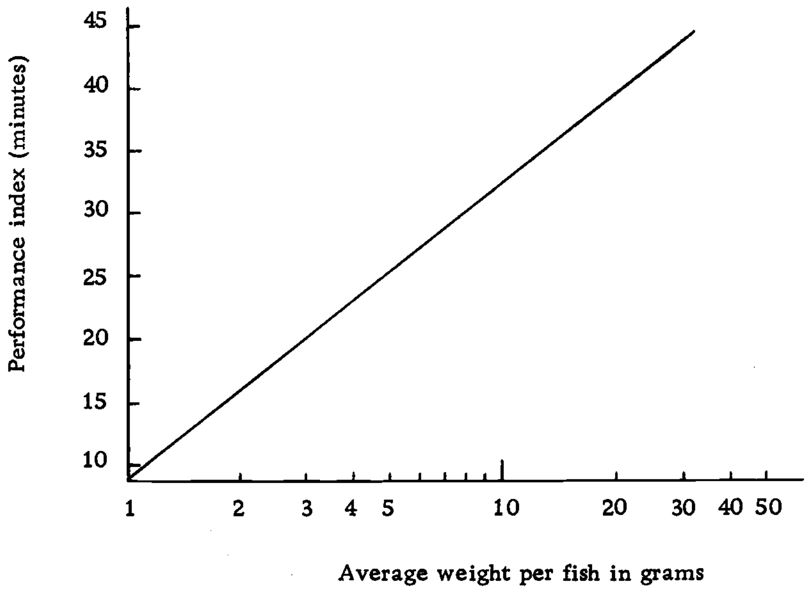

Figure 14. Graph depicting increase in performance with increase in weight in fingerling fall chinook salmon. (Modified from Thomas, Burrows and Chenoweth, 1964).

endurance than fish in fresh water or in 50 percent sea water tested at the same time due to the larger size of the latter fish. This agrees with experiments demonstrating the greater endurance of large salmon compared to that of small salmon (Bams, 1967; Brett, Hollands and Alderdice, 1958; Thomas, Burrows, and Chenoweth, 1964).

In Group I, the performance of salmon living in 100 percent sea water relative to weight was similar to the performance of the fish living in fresh water and in 50 percent sea water. The wt./p.i. ratios of salmon acclimated to 100 percent sea water tended to decrease as fish weight decreased (see May 27th and May 29th tests, Table 11); the same would be true for any population of salmon in fresh water.

The regression line for Group I fish (Figure 12) intercepts the fish weight-axis near 0. This line is meaningful since fish weighing 1.07 grams (log = .03) would have little or no endurance at the initial low water velocity used in endurance testing.

Group II salmon in 100 percent sea water demonstrated lower performance than fish in 50 percent sea water. The wt./p.i. ratios for the salmon living in 100 percent sea water were numerically larger than ratios for fish in 50 percent sea water even though the latter fish weighed more (see May 26th and May 28th tests, Table 12).

The fish weight-axis intercept of the regression line for Group II fish (Figure 13) is numerically large, since fish weighing around

Table 11. Endurance test results for Group I salmon using the Category II test schedule.

Subgroup	Salinity	Date tested	Performance index (minutes)	Mean weight (grams)	Wt./p.i. ratio	Swimming tunnel water temp. (°C) start	finish
Ia	0	May 27	37.1	4.96	.134	14.0	14.5
Ib	50	May 27	34.1	4.21	.124	14.0	14.5
Ic	100	May 27	28.6	2.90	.102	14.0	14.5
Ie	100	May 27	24.8	2.64	.106	13.5	14.0
Ia	0	May 29	36.7	5.08	.142	14.3	14.8
Ib	50	May 29	31.6	4.29	.136	14.0	14.5
Ic	100	May 29	21.3	2.81	.132	13.5	14.0
Ie	100	May 29	19.7	2.59	.132	13.5	14.0
Ia	0	June 4	31.6	5.73	.148	15.6	16.0
Ic	100	June 4	18.9	3.01	.160	11.5	12.0
Ie	100	June 4	13.1	2.68	.204	12.0	12.5

Table 12. Endurance test results for Group II salmon using the Category II test schedule.

Subgroup	Salinity	Date tested	Performance index (minutes)	Mean weight (grams)	Wt./p.i. ratio	Swimming tunnel water temp. (°C) start	finish
IIe	50	May 26	35.2	2.53	.072	14.0	14.0
IIh	100	May 26	24.6	2.16	.088	12.5	13.0
IIf	100	May 26	22.7	1.89	.084	13.0	12.5
IIc	100	May 26	16.4	1.78	.108	12.0	13.5
IIe	50	May 28	30.3	2.90	.096	13.5	14.0
IIh	100	May 28	21.0	2.16	.104	13.8	14.1
IIf	100	May 28	21.7	2.10	.096	13.0	13.5
IIc[1/]	100	May 28	27.1	1.85	.068	14.8	15.0
IIe	50	June 2	25.3	3.03	.100	13.0	13.0
IIh	100	June 2	9.2	2.31	.230	10.0	10.5
IIg	100	June 2	8.8	2.04	.252	10.0	10.3

[1/] The May 28th test data for this subgroup were not used in determining the regression line in Figure 13 because the water was electrically charged during part of the test.

1.3 grams (log = .11) would probably have a performance index greater than 0.

Temperature probably played a role in the low performance of salmon in 100 percent sea water tested on May 26th (Table 12). This will be discussed in more detail in the section dealing with the effect of salinity on endurance.

All Group II subgroups performed better than their Group I counterparts as indicated by their smaller wt./p.i. ratios relative to weight.

The regression line slope for the Group II subgroups (Figure 13) is numerically less than the slope for Group I fish suggesting the possibility that Group II fish had a greater increase in performance per unit increase in weight than Group I fish. A Student's "t" test for the difference between the slopes of the two regression lines was conducted using the same equations listed on page 31. There was no statistical difference between the slopes ($p > 0.01$).

Two stamina tests were conducted, using the Category II endurance schedule, to demonstrate the debilitating effect of low temperatures on fish performance. Rearing-tank temperatures were equal to swimming-tunnel temperatures for four days prior to testing. In Group I, two subgroups (Ic and Ie) in 100 percent sea water were tested at temperatures which were 3.6 C to 4.1 C lower than the fresh-water temperature (see June tests, Table 11). Even though the fish in fresh water weighed at least 2.5 grams more than those in 100 percent sea water, the wt./p.i. ratios were much larger for the latter fish, indicating their poor performance at the lower temperatures. A similar test with Group II fish indicated that fish in 100 percent sea water which weighed less than and were tested at a 3.0 C lower temperature than the subgroup in 50 percent sea water had wt./p.i. ratios more than twice as large as those of the fish in 50 percent sea water (see June tests, Table 12).

Effect of Salinity on Endurance

Chinook salmon acclimated to 100 percent sea water early in their juvenile life would not necessarily be expected to demonstrate stamina equal or superior to that of fish in fresh water or in 50 percent sea water. Nevertheless, Group I salmon in 100 percent sea water were able to swim against a known current as well as salmon of the same size reared in fresh water. Furthermore, the Group II fish in 50 and 100 percent sea water appeared to have superior stamina relative to their weight than the Group I fish.

The mode of acclimation to 100 percent sea water appeared to have little or no bearing on the stamina of the fish. Fish of the same size, whether placed directly into 100 percent sea water from 50 percent or transferred first to an intermediate salinity (75 percent), exhibited a similar capability to swim against a known current.

Fish endurance can be affected by a number of factors in addition to salinity including water temperature, oxygen concentration, food intake, and crowding.

1. Temperature plays an important role in fish endurance, with fish performance increasing as the temperature increases within a given optimal range (Brett, Hollands and Alderdice, 1958; Davis et al., 1963; Thomas, Burrows and Chenoweth, 1964). This increase in endurance is a result of the effect of temperature on salmon physiology and water viscosity (Thomas, Burrows and Chenoweth, 1964).

In my tests, salmon in 100 percent sea water tested at temperatures ranging from 3.0 C to 4.0 C lower than for fish in fresh water demonstrated relatively poor performance (see June tests, Tables 11 and 12).

A week prior to endurance testing, 100 percent sea-water temperatures averaged about 3.0 C and 1.0 C lower than fresh-water and 50 percent sea-water temperatures, respectively. All water temperatures were within 2.0 C during the four days of stamina testing under the Category II endurance schedule (Tables 11 and 12). The poor performance of Group II salmon acclimated to 100 percent sea water in comparison to fish in 50 percent sea water on May 26 may be partly explained by lower water temperature prior to testing.

2. Oxygen concentration is another factor which bears

importantly on fish stamina. Davis *et al.* (1963) demonstrated that chinook salmon performance decreased as oxygen levels declined from 7.0 to 3.0 mg/l. Throughout all of my endurance tests, the oxygen concentrations were well above 8.0 mg/l and were considered as unchanging.

3. Hunger was also considered as a variable to be controlled in stamina testing. My test fish were unfed for at least 24 hours before testing because the energy needed for food digestion and assimilation can materially affect fish performance (Thomas, Burrows and Chenoweth, 1964). A starvation period of up to 32 hours did not appear to affect the performance of test fish.

4. Crowding was avoided in my stamina tests by limiting the maximum number of fish tested at a time to 12 per tunnel. Although the number of fish tested per tunnel was held constant for a particular test, total biomass varied according to the average size of the fish tested. In no case did the biomass of fish to volume of water ratio exceed 2.75 g/l.

DISCUSSION AND CONCLUSION

A 0-50-100 percent sea-water acclimation route for juvenile fall chinook salmon would be highly advantageous for the fish and the experimenter. The salmon would be placed in 50 percent sea water 40 days after hatching and then transferred to 100 percent sea water 90 days after hatching. Salmon should be as large as possible at the time of exposure to 100 percent sea water to facilitate growth and survival. This requires initial feeding at an early age. Salt and fresh water temperatures between $10^{o}C$ and $13^{o}C$ appear appropriate.

Compared to fish in fresh water and in 50 percent sea water, all fish acclimated to 100 percent sea water grew slower due to decreased food consumption and had lower absolute stamina due to smaller size. Stamina relative to size was not apparently affected by osmoregulatory stress.

The 50-100 percent sea-water acclimation route was more advantageous than the 50-75-100 percent sea-water route. Fish in 75 percent sea water grew slower than fish in 50 percent sea water. At the end of the experimental period, fish acclimated to 100 percent sea water via the 50-75-100 percent sea-water route were smaller and suffered higher mortality than fish acclimated via the 50-100 percent sea-water route.

Prediction of the success of ocean-released salmon acclimated to 100 percent sea water is difficult without further research. Salmon that survive for 14 days after exposure to full-strength sea water will likely survive ocean salinities. As the fish grow larger after ocean release, osmoregulatory stress will diminish.

Predation will be the major problem confronting the ocean released salmon. Fish acclimated to 100 percent sea water and then released in the ocean will be smaller than fish which normally migrate to the ocean from fresh water. Fish size at time of release into rivers seems to play a significant role in returns, with fingerling returns generally greater than fry returns (Cope and Slater, 1957; Junge and Phinney, 1963). This may be due to the ease with which predators can catch smaller fish.

In the ocean, predation may affect the smaller salmon acclimated to 100 percent sea water more than the larger salmon reared as juveniles in fresh water.

The problem of ocean predation on acclimated fish must be weighed against the problems confronting only salmon reared in fresh water. Ocean-released salmon would not be subjected to predation losses in fresh water, fresh-water pollution, dams, and fishing pressure on outgoing migrants.

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