

THERMAL TOLERANCES  
OF INTERIOR ALASKAN  
ARCTIC GRAYLING

A stylized, minimalist illustration of a river or stream. The water is represented by a dark, solid horizontal band across the upper middle. Below this, the riverbed is depicted with large, irregular, light-colored shapes outlined in dark lines, suggesting rocks or boulders. The overall style is graphic and modern.

INSTITUTE OF WATER RESOURCES

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THERMAL TOLERANCES OF INTERIOR ALASKAN

ARCTIC GRAYLING

*(Thymallus arcticus)*

Thermal tolerances of Interior Alaskan Arctic Grayling (*thymallus arcticus*)

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## INTRODUCTION

The arctic grayling (*Thymallus arcticus*, frontispiece) is a member of a family Salmonidae which also includes the salmon, trout, char and whitefishes. All are considered to be cold water fishes, that is, occurring naturally in waters that do not exceed 70°F. The former range of this fish in America included the cold water streams to the west of Hudson Bay with isolated populations found in Michigan and Montana (Eddy, 1957; Davis, 1970).

Because of its high value as a sports fish, this fish has been artificially propagated for introduction to streams where it does not naturally occur. During these hatchery efforts this fish has been found to be quite eurythermic and somewhat resistant to low dissolved oxygen levels related to high water temperatures (Davis, 1970).

These fish became extinct in the Michigan streams where they had formerly occurred about 1930. Vincent (1962) attributes this disappearance to three factors: logging, the scouring of spawning areas by floating logs, and competition with stocked rainbow trout. One of the most serious effects of logging along trout streams has been the reduction of shading and the associated rise in water temperatures (Iwanaga and Hall, 1973). Questions then arise concerning the ultimate effects of thermal loading on natural populations of arctic grayling.

The population of grayling that was the focus of this project spawns and summers in the Chena River in Interior Alaska providing a popular



sports fishery for residents and visitors to the area. General information concerning this population and those of nearby streams is available in unpublished theses by Wojcik (1955), and Vascotto (1970).

This population is currently exposed to thermal pollution along the lower stretches of the Chena (Fig. 1) where two sewage treatment plants and a conventional power plant are located. There the river has a naturally wide bed and gentle slope and the thermal additions, relatively high in volume, are sufficient to keep several miles of water open through the severe subarctic winter. The fish spawn and summer upstream from this area and run through it downstream in the fall to spend the winter in the glacial Tanana River which is clear-running in winter. Thus the grayling in the Chena River are perhaps little affected by the thermal loading presently entering the stream.

A flood control project to protect the city of Fairbanks is scheduled for immediate construction. The project site is about 17 air miles from the present area of thermal additions and lies within the summer range of this population although probably downstream from prime spawning beds. A fish ladder is incorporated into the project design and is planned to allow the spring and fall runs of grayling to continue as well as fall runs of salmon. The drawdown of the two shallow reservoirs impounded between the Chena and Tanana Rivers is expected to increase the river temperature by one or two degrees Centigrade as a theoretical maximum. Thus, among other effects this project will have on the environment of these grayling will be thermal loading in an area that is presently free of pollution of any type. The question then arises whether the effects of this thermal loading, if any, can be minimized by proper management of the project. First, life cycle stages sensitive to thermal effects must be identified.

The experiments described in this report were conducted as standard 96-hour bioassays (A.P.H.A. 1970). In this type of test the pollutant

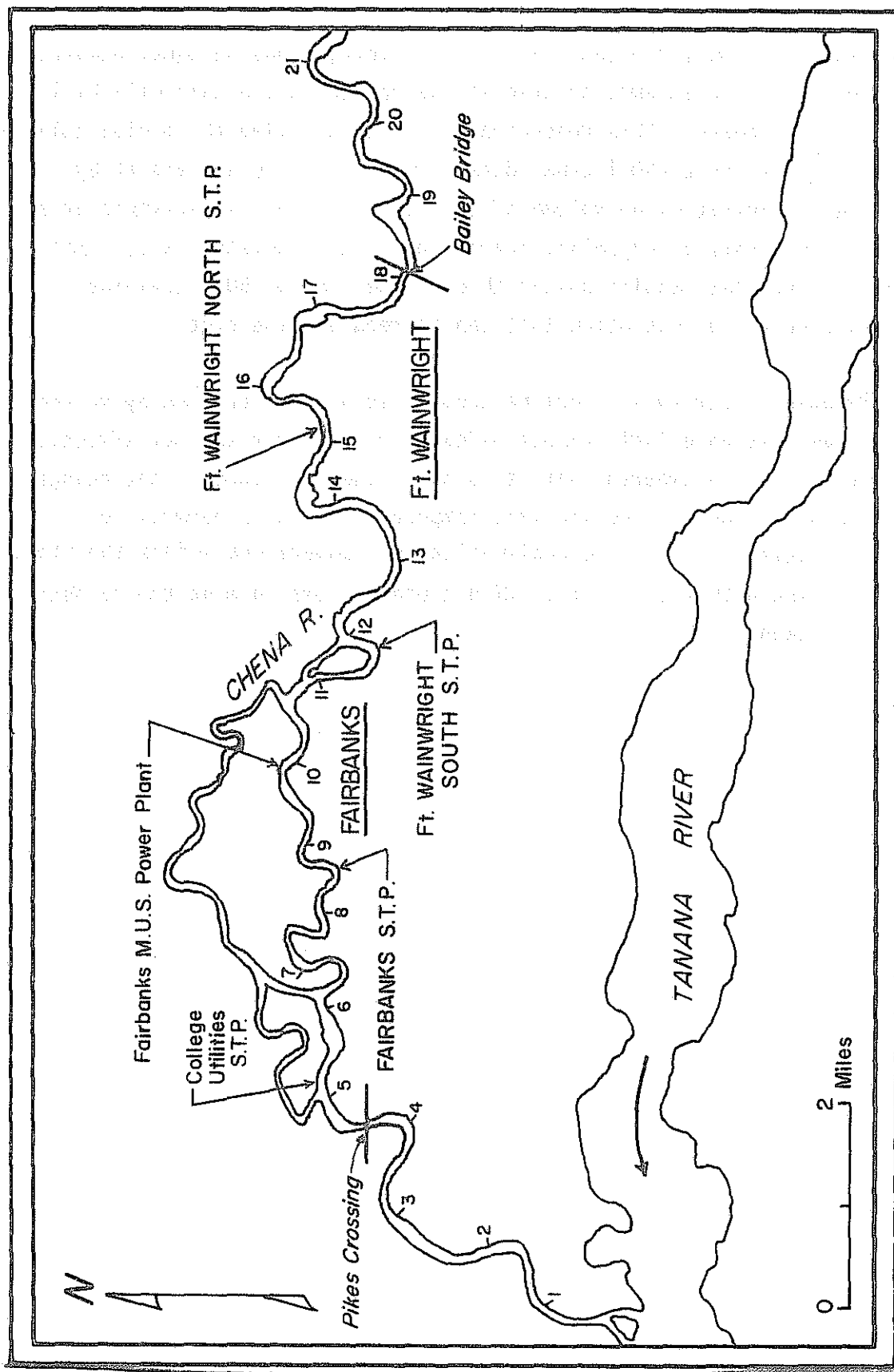


Fig. 1: Sources of thermal pollution along the lower Chena River  
(S.T.P. indicates sewage treatment plant)

is tested in as many concentrations as possible, exposing equal numbers of live fish in an attempt to bracket the concentration that will kill 50% of fish exposed. This concentration is then called the median tolerance limit ( $TL_m$ ) or the LD-50 (lethal dose for 50%), and is arrived at by plotting concentration by volume of the pollutant on a logarithmic axis against the number of organisms surviving on an arithmetic axis. The straight line that results passes through the line of 50% surviving at a particular concentration that can be read off the plot.

Because temperature cannot be considered a concentration by volume, the median tolerance limit cannot be exactly fixed for thermal effects. It must also be remembered that it is often the magnitude of the change in temperature and not the specific temperature that is harmful to living organisms. Thus, the acclimatization temperature before the bioassay must be noted with the results. With these factors in mind the project was undertaken.

## METHODS

### *OBTAINING THE FISH*

The original plan for providing the fish for this project called for the capture of mature fish during the 1972 spawning run on the Chena River by Alaska Department of Fish and Game personnel. Some of these fish were to be tested in the experimental setup and the remaining were to be artificially spawned. Thus obtained, fertilized eggs were to be hatched at the Fire Lake Hatchery (ADF&G) near Anchorage, Alaska, and air-freighted to the laboratories of the Institute of Water Resources immediately after hatching and thereafter at five centimeter increments of growth. This plan had to be modified due to the catastrophic death of all remaining grayling fry at the hatchery shortly after the first shipment of sac fry to IWR laboratories was completed a few days after hatching.

Fairbanks ADF&G personnel subsequently captured mixed sized fish from the Chena River by electric shocking methods and transported fish of as uniform size as possible to the Institute. Young of the year were obtained from the river by netting while they were schooling. The fish captured from the river were brought to the laboratories by truck in containers filled with river water.

### *WATER*

Initially it was assumed that tap water could be used for holding and experimental purposes after it had been allowed to stand for several

days to allow the chlorine to escape. The first set of mature fish returned to the laboratories were placed in a five hundred gallon refrigerated circular tank containing tap water that had stood for approximately eight days and showed no chlorine content when tested by the Hach Test Kit method. However, approximately four hours later when the fish were checked, the majority of them were in distress. All of the fish succumbed before the next morning. The cause of the distress was found to be high pH (about 9.0).

For the remainder of the project, water for the holding and experimental tanks was transported from the Chena River on the U.S. Government property near Fort Wainwright above all major outfalls that enter the lower Chena River. The water was hauled in glass or plastic containers that had been rinsed with 10% hydrochloric acid cleaning solution.

#### *TANKS*

For the most part, fish were held prior to testing in a 500 gal. (1890 liter) refrigerated tank molded of fiber glass. Refrigeration was provided by a long-shafted Frigid Systems, Inc., unit. This unit had an upper temperature limit of about 10°C depending somewhat on the ambient temperature.

Experiments were primarily conducted in four 200 gal. (756 liter) rectangular fiber glass tanks called Living Streams cooled by Frigid Systems, Inc., units. These tanks could be controlled within plus or minus one degree between 5°C and 20°C. Fish that were too small to run free in these tanks were isolated in 5 gal. (18.9 liter) bioassay jars for protection from the cooling system impellor.

Some of the young of the year were tested in 3 gal. (11.34 liter) battery jars placed in a New Brunswick Scientific Co., Inc., PsychroTherm

Controlled Environment Incubator Shaker which allowed these fish to be tested at temperatures above 20°C with good temperature control.

The final experiment was conducted with mature and nearly mature fish in two new 50 gal. (189 liter) Instant Ocean tanks which had been factory-modified to give plus or minus  $\frac{1}{2}$  degree control between 20°C and 25°C.

#### *TRANSFER*

Fish were transferred from the circular 500 gal. holding tank to the experimental tanks using fine-meshed hand nets. Often the length of each fish was measured at this time using a plastic fish-measuring board.

When the fish were small enough to require protection from the refrigerator impellor, the fish were usually placed into bioassay jars before being placed in the holding tank. At the initiation of an experiment, the jars were transferred to the experimental tanks thus subjecting these smaller fish initially to a gradual change in temperature as it took several hours for the water in the jar to reach equilibrium with the water surrounding it in the experimental tank.

#### *CONTROLS*

Fish were held as long as practical in the laboratory so that the effects of capture and transport to the laboratory would subside and the fish would become acclimatized to the laboratory situation.

Any fish not transferred to an experimental tank from the holding tank were considered to be controls for the experiment. In some experiments, the coolest experimental tank was near the temperature of the

holding tank and these fish can be considered more proper controls as they also underwent the stress of being transferred to a different tank.

#### *FEED*

In accordance with the standard methods recommended for bioassays (A.P.H.A. 1971), feeding of the fish, if any, was usually discontinued 48 hours before an experiment was begun and not resumed until termination of the bioassay.

The foods presented to the larger grayling were freeze-dried brine shrimp nauplii and cooked liver. Feeding activity was found to be more intense when liver was presented.

Freshly hatched brine shrimp nauplii were presented to the sac fry grayling about four days after their arrival at the laboratory and throughout the experiment to prevent starvation. Feeding success was not observed, however, until the termination of the experiment on this size class at which time the fish were ten days old. Brine shrimp nauplii were then observed only in the guts of sac fry that had been exposed to 15°C and 20°C water. This indicates that higher water temperature increased the speed of metabolism of the yolk sac triggering early feeding success.

The last group of fish were held about two weeks while the water for the experimental tanks was being hauled and the tanks conditioned. These fish were fed mosquito larvae and pupae netted from temporary standing water.

#### *OXYGEN*

Aeration was provided to all tanks and jars by piped compressed air distributed through airstones. When small fish were assayed in battery jars these airstones were contained in aquarium filters packed with fiber



glass. Oxygen levels were monitored once every eight hours in several of the first experiments. Water was removed from the Living Streams with a polyvinyl chloride miniaturized dissolved oxygen sampler. The water was analyzed for dissolved oxygen using Hach powder pillows for fixation and the Hach burette for titration. In the tests conducted with the smaller fish isolated in bioassay jars, the Hach method could not be used as it required about 330 ml of water to be removed each time the test was conducted, and additionally, the sampler would have disturbed the fish too much. A dissolved oxygen probe, which uses current produced in a redox reaction, was planned for use in limited amounts of water. This method never proved to be possible due to the failure of all probes tested.

#### *TEMPERATURE*

During experiments one through four, temperature of the water was measured with a mercury-filled Centigrade thermometer once every eight hours when the fish were checked. During the remaining experiments, temperature was monitored throughout the test with a 24 point Honeywell Pyrometer Recorder. Thermocouple wires were immersed in each tank, and water temperature was printed on a paper record.

## RESULTS

EXPERIMENT #1, MAY 23, 1972:

TABLE 1.  
A RECIRCULATED FLOW BIOASSAY OF THERMAL EFFECTS  
ON *Thymallus arcticus* DURING 96 HOURS  
(5/23/72 - 5/27/72)

Holding Conditions: 200 gal. rectangular tank; 4°C ± 1°C; 6 days  
5/17/72 - 5/23/72; feed - none.

Control Fish: 3\*; mean size 17.9 cm

| TANKS                         | 200 gal | 200 gal | 200 gal | 200 gal |
|-------------------------------|---------|---------|---------|---------|
| DESIRED TEMPERATURE           | 5°C     | 10°C    | 15°C    | 20°C    |
| HIGHEST TEMPERATURE           | 5.4°C   | 10.5°C  | 17.0°C  | 20.0°C  |
| LOWEST TEMPERATURE            | 4.0°C   | 9.6°C   | 15.9°C  | 18.5°C  |
| FISH: MEAN SIZE               | 17.9 cm | 20.2 cm | 21.9 cm | 23.8 cm |
| NO. FISH TRANSFERRED          | 3*      | 8       | 10      | 10      |
| NO. SURVIVING 96 HOURS        | 3       | 6       | 10      | 10      |
| NO. DEAD EXPERIMENTAL EFFECTS | 0       | 0       | 0       | 0       |
| NO. DEAD OTHER REASONS        | 0       | 2       | 0       | 0       |

\*These fish were left in the holding tank.

## REMARKS

Fish placed into the 20° tank first reacted by random searching. After about 24 hours, they would strike at the thermometer when it was immersed in the water.

Some eggs were found in the 10°C tank during cleaning of tanks at termination. Some of these fish seemed to exhibit spawning behavior when first transferred to the 10°C tank from the holding tank. When returned to the control-holding tank, the 20°C tested fish were very sluggish at first, grouping together, and one or two were nosing the surface.

In this and all other experiments a tally is kept of the number of fish dead from other reasons than experimental effects. These other reasons include such events as fish jumping out of the tank, fish entering the impellor area of the tank past a loose screen, and the like.

All live fish were returned to the Chena River and released.

EXPERIMENT #2, MAY 30, 1972:

TABLE 2.  
A RECIRCULATED FLOW BIOASSAY OF THERMAL EFFECTS  
ON *Thymallus arcticus* DURING 96 HOURS  
(5/30/72 - 6/2/72)

Holding Conditions: 500 gal. circular tank;  $4^{\circ}\text{C} \pm 1^{\circ}\text{C}$ ; 5 days  
5/25/72 - 5/30/72; feed - none

Control Fish: 20; mean size 18.6 cm

|                               |                       |                        |                        |                        |
|-------------------------------|-----------------------|------------------------|------------------------|------------------------|
| TANKS                         | 200 gal               | 200 gal                | 200 gal                | 200 gal                |
| DESIRED TEMPERATURE           | $5^{\circ}\text{C}$   | $10^{\circ}\text{C}$   | $15^{\circ}\text{C}$   | $20^{\circ}\text{C}$   |
| HIGHEST TEMPERATURE           | $5.2^{\circ}\text{C}$ | $10.2^{\circ}\text{C}$ | $16.4^{\circ}\text{C}$ | $20.0^{\circ}\text{C}$ |
| LOWEST TEMPERATURE            | $4.5^{\circ}\text{C}$ | $9.0^{\circ}\text{C}$  | $14.0^{\circ}\text{C}$ | $19.3^{\circ}\text{C}$ |
| FISH: MEAN SIZE               | 18.6 cm               | 18.8 cm                | 20.5 cm                | 20.6 cm                |
| NO. FISH TRANSFERRED          | 20                    | 15                     | 15                     | 15                     |
| NO. SURVIVING 96 HOURS        | 19                    | 15                     | 15                     | 15                     |
| NO. DEAD EXPERIMENTAL EFFECTS | 0                     | 0                      | 0                      | 0                      |
| NO. DEAD OTHER REASONS        | 1                     | 0                      | 0                      | 0                      |

#### REMARKS

The fish that died had been dropped on the floor and appeared after death to have a broken backbone. After 24 hours the fish in the  $20^{\circ}\text{C}$  tank were very excitable, but this irritability subsided by the termination of the experiment.

All live fish were returned to the Chena River and released.

EXPERIMENT #3, JUNE 2, 1972:

TABLE 3.  
A STATIC BIOASSAY OF THERMAL EFFECTS  
ON *Thymallus arcticus* DURING 18 HOURS  
(6/2/72 - 6/3/72)

Holding Conditions: 500 gal. circular tank;  $3^{\circ}\text{C} \pm 1^{\circ}\text{C}$ ; 2 days  
5/30/72 - 6/2/72; feed - none.

Control Fish: 9\*; mean size 12.2 cm.

---

TANKS: A 400 gal. square metal tank sealed with aquarium paint. This tank had no refrigeration unit nor thermostat. The water was heated slightly above ambient with a lightbulb placed under the center of the tank.

---

|                               |          |
|-------------------------------|----------|
| HIGHEST TEMPERATURE           | 25.2°C   |
| LOWEST TEMPERATURE            | 25.0°C   |
| MEAN SIZE                     | 17.97 cm |
| NO. FISH TRANSFERRED          | 66       |
| NO. SURVIVING 18 HOURS        | 0        |
| NO. DEAD EXPERIMENTAL EFFECTS | 66       |
| NO. DEAD OTHER REASONS        | 0        |

---

\*These fish were left in the holding tank.

#### REMARKS

Fifteen fish died during the first hour and an additional 45 were dead within  $6\frac{1}{2}$  hours of the beginning of the test. The water in the tank was suspected to be a possible factor in the mortality because of a musty aromatic odor. Bioassay jars of this water cooled in a refrigerated tank were tested, however, with live fish and no mortality ensued.

All control fish survived and were released.

EXPERIMENT #4, JUNE 27, 1972:

TABLE 4.

A RECIRCULATED FLOW BIOASSAY OF THERMAL EFFECTS  
ON *Thymallus arcticus* DURING 96 HOURS  
(6/27/72 - 7/1/72)

Holding Conditions: 500 gal. circular tank; 7°C ± 1°C; 6 days  
6/21/72 - 6/27/72; feed - none.

Control Fish: None

|                                  |         |         |         |         |
|----------------------------------|---------|---------|---------|---------|
| TANKS                            | 200 gal | 200 gal | 200 gal | 200 gal |
| DESIRED TEMPERATURE              | 20°C    | 18°C    | 20°C    | 20°C    |
| HIGHEST TEMPERATURE              | 20.4°C  | 17.7°C  | 24.6°C  | 22.0°C  |
| LOWEST TEMPERATURE               | 19.2°C  | 15.8°C  | 20.2°C  | 20.3°C  |
| FISH: MEAN SIZE                  | 18.4 cm | 18.1 cm | 16.6 cm | 17.3 cm |
| NO. TRANSFERRED                  | 35      | 35      | 35      | 35      |
| NO. SURVIVING 96 HOURS           | 33      | 32      | 2       | 30      |
| NO. DEAD EXPERIMENTAL<br>EFFECTS | 0       | 0       | 0       | 0       |
| NO. DEAD OTHER REASONS           | 2       | 3       | 33      | 5       |

REMARKS

This experiment should have been started in fresh water. Instead, water used in previous testing was used. Because of turbidity, it was necessary to pump the water in all of the tanks through a glass wool filter in mid-test. Several fish were observed alive in the #2 20°C tank at 24.6°C. This temperature, reached about 56 hours after the test was begun, was probably due to heat produced by the submersible pump used to filter the water. During those first 56 hours, about half of the fish in this tank were killed by the impellor because of a loose screen.

Of the approximately 15 not killed by the impellor, all but two died as a consequence of a low dissolved oxygen concentration resulting from ground-up fish in the tank. An alternative possibility, of course, remains that death from thermal stress occurred.

All live fish were returned to the Chena River and released.



EXPERIMENT #5, JULY 8, 1972:

TABLE 5.

A STATIC BIOASSAY OF THERMAL EFFECTS  
ON *Thymallus arcticus* SAC FRY DURING 96 HOURS  
(7/8/72 - 7/11/72)

Holding Conditions: 12,500 placed into each of two 5 gal. bioassay jars and immersed in the water of the 500 gal. circular tanks;  $4^{\circ}\text{C} \pm 1^{\circ}\text{C}$ ; 6 days 7/2/72 - 7/8/72; feed - brine shrimp nauplii.

Control Fish: Uncounted thousands remaining in bioassay jars; mean size unknown.

|                               |       |        |        |        |
|-------------------------------|-------|--------|--------|--------|
| TANKS                         | 5 gal | 5 gal. | 5 gal. | 5 gal. |
| DESIRED TEMPERATURE           | 5°C   | 10°C   | 15°C   | 20°C   |
| HIGHEST TEMPERATURE           | 5.0°C | 9.0°C  | 16.3°C | 21.2°C |
| LOWEST TEMPERATURE            | 2.8°C | 7.5°C  | 14.6°C | 19.7°C |
| FISH: MEAN SIZE               | 14 mm |        |        |        |
| NO. FISH TRANSFERRED          | 473   | 578    | 307    | 177    |
| NO. SURVIVING 96 HOURS        | 446   | 549    | 294    | 171    |
| NO. DEAD EXPERIMENTAL EFFECTS | 27    | 16     | 12     | 6      |
| NO. DEAD OTHER REASONS        | 0     | 13     | 1      | 0      |

REMARKS

The sac fry developed a fungus problem which was treated with formalin before testing began. The best way found to treat this fungus was to add approximately 3 mililiters of formalin from a wash bottle to about 3 gallons of water containing the sac fry. Live fish swam up from the fungal mat and were netted off and placed in fresh water.

These fish were fed throughout the test and holding periods. The fish tested in the 15°C and 20°C tanks had used up their yolk sacs by the end of the test; brine shrimp eggs and nauplii were found in their intestinal tracts. All fish used in the experiment were sacrificed for counting. The remaining live control fish were used in further experiments.

EXPERIMENT #6, JULY 19, 1972:

TABLE 6.  
A STATIC BIOASSAY OF THERMAL EFFECTS  
ON *Thymallus arcticus* SAC FRY DURING 16 HOURS  
(7/19/72 - 7/20/72)

Holding Conditions: Several thousand in each of two 5 gal. bioassay jars immersed in the water of the 500 gal. circular tank;  $5^{\circ}\text{C} \pm 3^{\circ}\text{C}$ ; 17 days 7/2/72 - 7/19/72; feed - brine shrimp nauplii.

Control Fish: Those left in the bioassay jars in the holding tank; mean size about 14 mm.

|                               |                        |                        |
|-------------------------------|------------------------|------------------------|
| TANKS                         | 3 gal                  | 3 gal                  |
| DESIRED TEMPERATURE           | $24^{\circ}\text{C}$   | $24^{\circ}\text{C}$   |
| HIGHEST TEMPERATURE           | $24.2^{\circ}\text{C}$ | $24.2^{\circ}\text{C}$ |
| LOWEST TEMPERATURE            | $8.5^{\circ}\text{C}$  | $8.5^{\circ}\text{C}$  |
| FISH: MEAN SIZE               | 14 mm (7/14/72)        |                        |
| NO. TRANSFERRED               | 129                    | 201                    |
| NO. SURVIVING 16 HOURS        | 0                      | 0                      |
| NO. DEAD EXPERIMENTAL EFFECTS | 129                    | 201                    |
| NO. DEAD OTHER REASONS        | 0                      | 0                      |

#### REMARKS

This experiment was conducted as a standard static bioassay in battery jars in an incubator without aeration. The fish were transferred to battery jars filled with water which was at the temperature of the holding tank  $8.5^{\circ}\text{C}$  to prevent shock. About one-fourth of the fry were dead in each of two battery jars at 9:45 p.m., July 19, 5 hours and 5 minutes after the test began. The water temperature at that time was

21.5°C. All fry were dead by 9:00 a.m. the next day, July 20, at which time the water temperature had reached 24.2°C and was holding steady. On July 23, all control sac fry being held in the holding tank in bioassay jars were dead. Some were still on their yolk sacs after 21 days.

This experiment was planned again when it could be more closely observed, but this was made impossible by the catastrophic death of all the sac fry being held in the holding tank. The cause of the catastrophic death of all the sac fry in the lab on July 23 is unknown.

EXPERIMENT #7, AUGUST 7, 1972:

TABLE 7.

A STATIC BIOASSAY OF THERMAL EFFECTS  
ON *Thymallus arcticus* YOUNG OF THE YEAR DURING 96 HOURS  
(8/7/72 - 8/10/72)

Holding Conditions: Four 5 gal. bioassay jars immersed in a 200 gal.  
rectangular tank;  $9^{\circ}\text{C} \pm 1^{\circ}\text{C}$ ; 6 days 8/1/72 -  
8/7/72; feed - freeze-dried brine shrimp

Control Fish: None

|                                  |                       |                       |                        |                        |                        |
|----------------------------------|-----------------------|-----------------------|------------------------|------------------------|------------------------|
| TANKS                            | 5 gal                 | 5 gal                 | 5 gal                  | 5 gal                  | 3 gal                  |
| DESIRED TEMPERATURE              | $5^{\circ}\text{C}$   | $10^{\circ}\text{C}$  | $15^{\circ}\text{C}$   | $20^{\circ}\text{C}$   | $25^{\circ}\text{C}$   |
| HIGHEST TEMPERATURE              | $6.8^{\circ}\text{C}$ | $9.3^{\circ}\text{C}$ | $17.1^{\circ}\text{C}$ | $20.3^{\circ}\text{C}$ | $25.3^{\circ}\text{C}$ |
| LOWEST TEMPERATURE               | $5.9^{\circ}\text{C}$ | $8.3^{\circ}\text{C}$ | $15.1^{\circ}\text{C}$ | $20.0^{\circ}\text{C}$ | $24.0^{\circ}\text{C}$ |
| FISH: MEAN SIZE                  | 5.5 cm (estimated)    |                       |                        |                        |                        |
| NO. FISH TRANSFERRED             | 52                    | 43                    | 35                     | 49                     | 21                     |
| NO. SURVIVING 96 HOURS           | 52                    | 41                    | 35                     | 48                     | 13                     |
| NO. DEAD EXPERIMENTAL<br>EFFECTS | 0                     | 2                     | 0                      | 1                      | 8                      |
| NO. DEAD OTHER REASONS           | 0                     | 0                     | 0                      | 0                      | 0                      |

REMARKS

All fish except those placed into the  $24^{\circ}\text{C}$  water were transferred in their bioassay jar from the holding tank to the experimental tank, thus the water temperature initially was about  $10^{\circ}\text{C}$  for all of these jars. The fish placed from  $10^{\circ}\text{C}$  water into water at  $24^{\circ}\text{C}$  acted shocked and excitable. Three died within the first seven hours, probably from shock, and two dead were found hidden under an aquarium filter in this jar at termination. The initial excitability and rapid ventilation of

fish in the 25°C water had subsided by the third day. The surviving fish behaved normally.

Of those placed into the 24°C water, a few were netted from each of the holding bioassay jars and placed directly into 24°C water in a battery jar in an incubator.

All remaining live fish were returned to the Chena River and released.

EXPERIMENT #8, AUGUST 28, 1972:

TABLE 8.  
A STATIC BIOASSAY OF THERMAL EFFECTS ON  
*Thymallus arcticus* YOUNG OF THE YEAR DURING 96 HOURS  
(8/28/72 - 8/31/72)

Holding Conditions: A 5 gal. bioassay jar immersed in a 200 gal. tank;  
10°C ± 1°C; feed - freeze-dried brine shrimp.

Control Fish: 12; mean size about 5.0 cm; 10°C ± 1°C.

|                               |                    |        |
|-------------------------------|--------------------|--------|
| TANKS                         | 3 gal              | 3 gal  |
| DESIRED TEMPERATURE           | 24.5°C             | 24.5°C |
| HIGHEST TEMPERATURE           | 24.5°C             | 24.5°C |
| LOWEST TEMPERATURE            | 24.5°C             | 24.5°C |
| FISH: MEAN SIZE               | 5.0 cm (estimated) |        |
| NO. FISH TRANSFERRED          | 17                 | 9      |
| NO. SURVIVING 96 HOURS        | 12                 | 8      |
| NO. DEAD EXPERIMENTAL EFFECTS | 5                  | 1      |
| NO. DEAD OTHER REASONS        | 0                  | 0      |

REMARKS:

The water in all of the jars, including the control jar, was 12°C at the start of the experiment and was cooled or warmed to the desired temperature.

The dead fish were found just before the termination of the experiment. All had been alive at 4 p.m. the previous day after 77½ hours of tests.

All live fish were sacrificed after the experiment.



EXPERIMENT #9, OCTOBER 16, 1972:

TABLE 9.

A STATIC BIOASSAY OF THERMAL EFFECTS ON  
*Oncorhynchus kisutch* (COHO SALMON) PARR DURING 96 HOURS  
(10/16/72 - 10/20/72)

Holding Conditions: Placed 100 in each of two 5 gal. bioassay jars in 200 gal. tank. Two days later transferred 50 from each into two more bioassay jars;  $10^{\circ}\text{C} \pm 1^{\circ}\text{C}$ ; 17 days 9/29/72 - 10/16/72; feed - freeze-dried brine shrimp and liver.

Control Fish: Less than 50; mean size approximately 60 cm;  $9^{\circ}\text{C} \pm 1^{\circ}\text{C}$ .

|                           |                             |                        |                        |                        |
|---------------------------|-----------------------------|------------------------|------------------------|------------------------|
| TANKS                     | 5 gal                       | 5 gal                  | 5 gal                  | 5 gal                  |
| DESIRED TEMPERATURE       | $10^{\circ}\text{C}$        | $15^{\circ}\text{C}$   | $20^{\circ}\text{C}$   | $30^{\circ}\text{C}$   |
| HIGHEST TEMPERATURE       | $9.0^{\circ}\text{C}$       | $17.0^{\circ}\text{C}$ | $20.0^{\circ}\text{C}$ | $26.2^{\circ}\text{C}$ |
| LOWEST TEMPERATURE        | $8.3^{\circ}\text{C}$       | $14.5^{\circ}\text{C}$ | $18.0^{\circ}\text{C}$ | $10.0^{\circ}\text{C}$ |
| FISH: MEAN SIZE           | 60 cm (estimated)           |                        |                        |                        |
| NO. TRANSFERRED           | Not Counted<br>Less than 50 | 37                     | 37                     | 36                     |
| NO. SURVIVING 96 HOURS    | Not Counted                 | 37                     | 37                     | 0                      |
| DEAD EXPERIMENTAL EFFECTS | 0                           | 0                      | 0                      | 36                     |
| DEAD OTHER REASONS        | 4                           | 0                      | 0                      | 0                      |

REMARKS

In this test all fish were transferred to bioassay jars with water at  $10^{\circ}\text{C}$ . Three of the jars were then placed into tanks set at higher temperatures. The fish were thus subjected to rising temperature for about the first hour-and-a-half to prevent shock. No fish in the warmest tank were dead at  $24.0^{\circ}\text{C}$  after 34 minutes, but all were dead at  $26.5^{\circ}\text{C}$  54 minutes

later. About 50% were dead at 26.2°C, and many of the rest were in turn-over.

All live fish were sacrificed after the experiment.

EXPERIMENT #10, JUNE 5, 1973

TABLE 10.  
A RECIRCULATED FLOW BIOASSAY OF THERMAL EFFECTS  
ON *Thymallus arcticus* DURING 96 HOURS  
(6/5/73 - 6/9/73)

Holding Conditions: 500 gal. circular tank;  $7^{\circ}\text{C} \pm 1^{\circ}\text{C}$ ; 13 days  
5/23/73 - 6/5/73; feed - mosquito larvae.

Control Fish: 12\* mean size 24.1 cm.

|                               |         |         |
|-------------------------------|---------|---------|
| TANKS                         | 50 gal  | 50 gal  |
| DESIRED TEMPERATURE           | 22.0°C  | 24.0°C  |
| HIGHEST TEMPERATURE           | 23.7°C  | 24.2°C  |
| LOWEST TEMPERATURE            | 21.5°C  | 24.0°C  |
| FISH: MEAN SIZE               | 24.4 cm | 23.9 cm |
| NO. FISH TRANSFERRED          | 10      | 10      |
| NO. SURVIVING 96 HOURS        | 7       | 0       |
| NO. DEAD EXPERIMENTAL EFFECTS | 0       | 9       |
| NO. DEAD OTHER REASONS        | 3       | 1       |

\*These fish were left in the holding tank.

#### REMARKS

The fish placed in the tank reacted immediately by losing equilibrium. Within ten minutes, three fish were in turn-over. All fish but two, one male and one female, were dead by 4:40 p.m., June 5. These two were found dead at 9:10 p.m. the same day, about seven hours after the test was begun.

The fish in the cooler (20°C) tank shed their external parasites and became agitated when humans approached on the afternoon of July 7, 1973. All survived until the termination of the test on June 9, 1973, but were in obviously weakened condition.

The 12 fish held as controls in the holding tank were released into the Chena River at the end of the 96 hour testing period. The remaining live fish were sacrificed at the advice of ADF&G personnel due to poor condition.

## SUMMARY

Nine bioassay experiments were conducted with *Thymallus arcticus*, the arctic grayling, at selected water temperatures with the objective of finding the median tolerance limit at the different life cycle stages. One experiment was conducted on fingerling Coho salmon *Oncorhynchus kisutch* to evaluate the experimental set-up using a fish for which the temperature tolerance is well-documented. The results of the experiment with silver salmon were well within the range reported by others. This indicated that proper equipment and techniques were being used.

TABLE 11.

RESULTS OF THERMAL BIOASSAYS ON *Thymallus arcticus*  
FROM THE CHENA RIVER NEAR FAIRBANKS, ALASKA

| Size of Fish      | Acclimatization Temperature | TL <sub>m</sub> Range |
|-------------------|-----------------------------|-----------------------|
| Sac Fry           | 8.5°C ± 1°C                 | 21.5°C - 24.2°C       |
| Young-of-the-year | 8.5°C ± 1°C                 | above 24.5°C          |
| >10 cm.           | 4°C ± 1°C                   | 20.0°C - 24.0°        |
| >20 cm.           | 8.0°C ± 1°C                 | 22.5°C - 24.5°C       |

In these experiments there was often as little as 2°C between a temperature that caused no mortality and the temperature that caused 100% mortality. This narrow difference is common with thermal bioassays on aquatic organisms (Mehursky and Kennedy, 1967). Brett (1960) in multivariate analysis on sockeye salmon thermal studies found that thermal loading

affected breeding success, growth, activity, and finally mortality as it was increased. His findings may be roughly applicable to other salmonid fishes including the grayling. This study did find, as in Brett's sockeye study, that mortality quickly intensified when temperature was raised only slightly above that temperature where mortality was first noted.

The results of this study are also typical of thermal bioassays as the temperature of median tolerance for that particular life cycle stage of the fish in question increases with acclimation temperature. This has been previously described by Mihursky and Kennedy (1967) in their state-of-the-art paper on water temperature criteria. Of course, there is an upper limit past which temperature fish cannot acclimate. This ultimate incipient lethal level was not identified in our experiments due to equipment limitations, especially in regard to the number of temperature-controlled tanks.

The variation in effect that temperature has on different life cycle stages of a particular organism was emphasized by Brett (1960) in reference to the paper by Mihursky and Kennedy (1967) and by Coutant (1969). This was found to be probably true for arctic grayling although it would require further experimentation to delineate all the differences.

Young of the year grayling acclimated at about 8°C did not suffer the immediate effects on equilibrium that grayling larger than 20 cm and acclimated at the same temperature suffered upon being transferred to 24°C water. In fact, the young of the year fish that survived for 96 hours at 24°C were acting completely normally at the termination of the test while the larger grayling were obviously in poor condition after the same span of time.

Further experimentation is recommended in this regard, especially at the egg stage of the grayling. This proved impossible in this study

because of the lack of incubation equipment. If further thermal bioassay experimentation is performed on the arctic grayling, it is recommended that some larger fish be sacrificed and checked for maturity, especially if the bioassays are run prior to and during breeding season. This is advisable to delineate the variation in response between juvenile fish larger than young of the year and mature adults. It is also recommended that some mature fish that have undergone the bioassay just prior to breeding season be stripped of their sex products if these develop normally and these products be tested for thermal effects on their viability. It is recognized that this would be a difficult undertaking as these fish would have to be collected prior to or during the breakup of the ice on the rivers they inhabit.

The most critical effect of thermal loading on the arctic grayling may well be the decreased ability of the water to solubilize oxygen. In most of these bioassays constant aeration was provided to eliminate low dissolved oxygen as a factor needing consideration. Figure 2 is a plot of dissolved oxygen concentration against temperature as measured in our first four experiments.

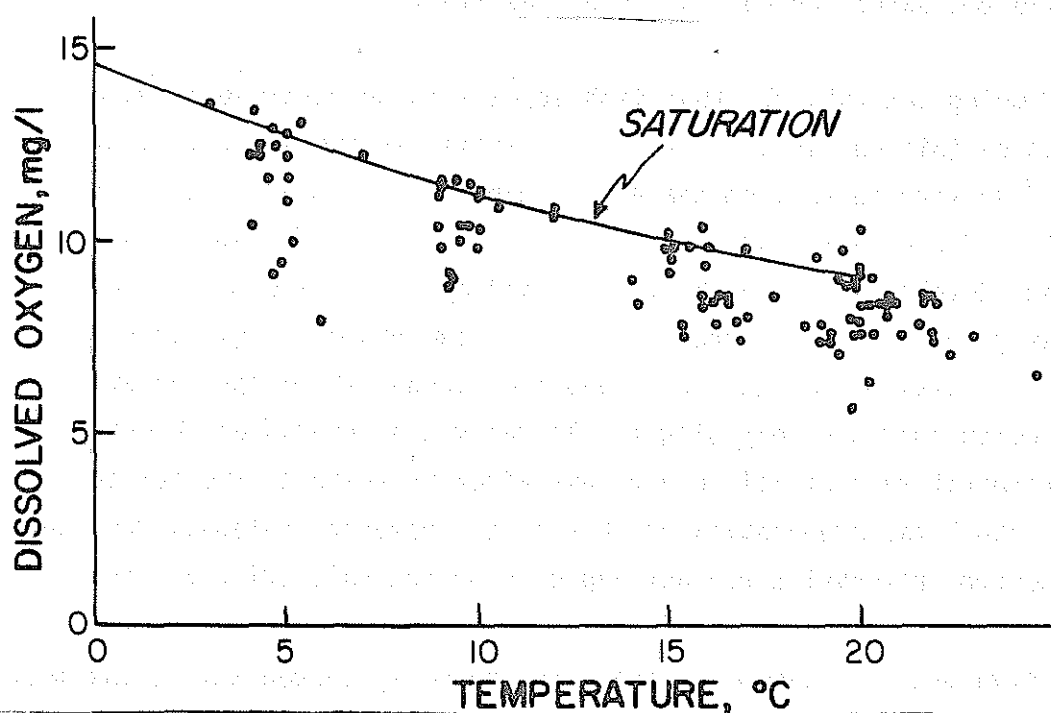


Fig. 2: Dissolved oxygen concentration in relation to temperature as measured in the first four experiments



The most severe decrease in dissolved oxygen concentration occurred once at 5.9°C when the dissolved oxygen concentration was only 8 mg/l. It should be noted that this concentration is almost 60% of the saturation concentration. It should also be noted that at no time during the bioassays when oxygen was being monitored was oxygen concentration found to be below 5 mg/l (the minimum concentration recommended for cold water fishes), nor was supersaturation a common occurrence. Thus, dissolved oxygen was assumed to be eliminated as a variable from these bioassay experiments.

Further study of the low temperature, low dissolved oxygen effects on the arctic grayling is recommended. These fish have been observed in the Chena River when the ice cover was complete and dissolved oxygen was measured near zero concentration (Roguski, 1972).

The slight elevation in water temperature expected from the flood control reservoirs to be constructed on the Chena River will probably have neither a direct effect on the grayling nor an indirect effect on the dissolved oxygen concentration of the river.

Spawning activity for this fish appears to be governed by water temperature (Bishop, 1971). This was verified in the laboratory when gravid fish were taken from the 4°C holding tank into an experimental tank at 10°C and some eggs were shed. It can also be assumed that final egg development is probably affected by a specific water temperature as grayling females are observed to have visible eggs almost immediately after breeding season and the changes these eggs undergo before being shed are very slight. Therefore, an unnatural change in water temperature just before and just after break-up of the ice cover could disturb the propagation of this fish. However, releases from the flood control reservoirs are not expected during this critical time.

Avoidance of stream areas that are thermally loaded may be the most important reaction of the fish. The Alaska Department of Fish and Game

has not captured any grayling during the winter where the stream is heated by the discharge of the Fairbanks Municipal Utilities power plant and the city sewage treatment plant (Tack, 1973). This may be due to insufficient methods of fishing but is more probably due to true avoidance of the area. This presents another question for further experimental treatment.

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