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Influence of Electrofishing on the Survival of Arctic Grayling, Chinook Salmon, Least Cisco, and Humpback Whitefish Eggs

by

Stafford M. Roach

April 1996

Alaska Department of Fish and Game

Division of Sport Fish



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| Weights and measures (metric) | | General | | Mathematics, statistics, fisheries | |
|---------------------------------------|--------------------|---|---|---|-------------------------|
| centimeter | cm | All commonly accepted abbreviations. | e.g., Mr., Mrs., a.m., p.m., etc. | alternate hypothesis | H_A |
| deciliter | dL | | | base of natural logarithm | e |
| gram | g | All commonly accepted professional titles. | e.g., Dr., Ph.D., R.N., etc. | catch per unit effort | CPUE |
| hectare | ha | and | & | coefficient of variation | CV |
| kilogram | kg | at | @ | common test statistics | F, t, χ^2 , etc. |
| kilometer | km | Compass directions: | | confidence interval | C.I. |
| liter | L | east | E | correlation coefficient | R (multiple) |
| meter | m | north | N | correlation coefficient | r (simple) |
| metric ton | mt | south | S | covariance | cov |
| milliliter | ml | west | W | degree (angular or temperature) | ° |
| millimeter | mm | Copyright | © | degrees of freedom | df |
| | | Corporate suffixes: | | divided by | ÷ or / (in equations) |
| | | Company | Co. | equals | = |
| | | Corporation | Corp. | expected value | E |
| | | Incorporated | Inc. | fork length | FL |
| | | Limited | Ltd. | greater than | > |
| | | et alii (and other people) | et al. | greater than or equal to | ≥ |
| | | et cetera (and so forth) | etc. | harvest per unit effort | HPUE |
| | | exempli gratia (for example) | e.g., | less than | < |
| | | id est (that is) | i.e., | less than or equal to | ≤ |
| | | latitude or longitude | lat. or long. | logarithm (natural) | ln |
| | | monetary symbols (U.S.) | \$, ¢ | logarithm (base 10) | log |
| | | months (tables and figures): first three letters | Jan,...,Dec | logarithm (specify base) | log ₂ , etc. |
| | | number (before a number) | # (e.g., #10) | mid-eye-to-fork | MEF |
| | | pounds (after a number) | # (e.g., 10#) | minute (angular) | ' |
| | | registered trademark | ® | multiplied by | x |
| | | trademark | ™ | not significant | NS |
| | | United States (adjective) | U.S. | null hypothesis | H_0 |
| | | United States of America (noun) | USA | percent | % |
| | | U.S. state and District of Columbia abbreviations | use two-letter abbreviations (e.g., AK, DC) | probability | P |
| | | | | probability of a type I error (rejection of the null hypothesis when true) | α |
| | | | | probability of a type II error (acceptance of the null hypothesis when false) | β |
| | | | | second (angular) | " |
| | | | | standard deviation | SD |
| | | | | standard error | SE |
| | | | | standard length | SL |
| | | | | total length | TL |
| | | | | variance | Var |
| Weights and measures (English) | | | | | |
| cubic feet per second | ft ³ /s | | | | |
| foot | ft | | | | |
| gallon | gal | | | | |
| inch | in | | | | |
| mile | mi | | | | |
| ounce | oz | | | | |
| pound | lb | | | | |
| quart | qt | | | | |
| yard | yd | | | | |
| Spell out acre and ton. | | | | | |
| Time and temperature | | | | | |
| day | d | | | | |
| degrees Celsius | °C | | | | |
| degrees Fahrenheit | °F | | | | |
| hour (spell out for 24-hour clock) | h | | | | |
| minute | min | | | | |
| second | s | | | | |
| Spell out year, month, and week. | | | | | |
| Physics and chemistry | | | | | |
| all atomic symbols | | | | | |
| alternating current | AC | | | | |
| ampere | A | | | | |
| calorie | cal | | | | |
| direct current | DC | | | | |
| hertz | Hz | | | | |
| horsepower | hp | | | | |
| hydrogen ion activity | pH | | | | |
| parts per million | ppm | | | | |
| parts per thousand | ppt, ‰ | | | | |
| volts | V | | | | |
| watts | W | | | | |

FISHERY MANUSCRIPT NO. 96-1

**INFLUENCE OF ELECTROFISHING ON THE SURVIVAL OF ARCTIC
GRAYLING, CHINOOK SALMON, LEAST CISCO, AND HUMPBACK
WHITEFISH EGGS**

by

Stafford M. Roach
Division of Sport Fish, Fairbanks

Alaska Department of Fish and Game
Division of Sport Fish, Research and Technical Services
333 Raspberry Road, Anchorage, Alaska, 99518-1599

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Stafford M. Roach

*Alaska Department of Fish and Game, Division of Sport Fish, Region III,
1300 College Road, Fairbanks, AK 99701-1599, USA*

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ABSTRACT

The influence of electrofishing on the survival of Arctic grayling *Thymallus arcticus*, chinook salmon *Oncorhynchus tshawytscha*, least cisco *Coregonus sardinella*, and humpback whitefish *C. sardinella* eggs was investigated. The hypothesis that electrofishing affects egg mortality was supported and egg mortality rates were discussed as a cost of obtaining biological information. Two experiments were conducted. Experiment 1 examined egg mortality rates for Arctic grayling, chinook salmon, least cisco, and humpback whitefish eggs from parents that were electroshocked, and Experiment 2 examined egg mortality rates for Arctic grayling and chinook salmon eggs electroshocked at various stages of development. For Experiment 1, when parents were electroshocked before spawning, egg mortality rates were significantly higher than control groups. Mean proportions of mortality rates increased 0.02 (SE = 0.01) for Arctic grayling eggs, 0.19 (SE = 0.01) for chinook salmon eggs, and 0.05 (SE = 0.02) for least cisco eggs compared to controls. The mean mortality rate for eggs from humpback whitefish parents that were electroshocked was 0.41 (SE = 0.01). Although not comparable to a control, this mortality rate was less than the mean mortality rate for either the control (0.52; SE = 0.01) or treated (0.57; SE = 0.01) eggs from least cisco parents. The eggs of least cisco and humpback whitefish are similar in size, rate of development, and ontogeny. For Experiment 2, when eggs were electroshocked after fertilization, Arctic grayling and chinook salmon mean egg mortality varied significantly by parents (electroshocked or not electroshocked), level of electroshock, and egg developmental stage. For Arctic grayling eggs, the largest difference in mean mortality rates compared to controls was 0.09 (SE = 0.05) for eggs electroshocked at the highest level (1.30 to 1.50 V/cm), at 70 temperature units post fertilization, and from parents that were electroshocked before spawning. For chinook salmon eggs, the largest difference in mean mortality rates compared to controls was 0.97 (SE = 0.03) for eggs electroshocked at the highest level, at 98 temperature units post fertilization, and from parents that were electroshocked before spawning. Under normal field conditions the high levels of electroshock would be experienced only by eggs that were in close proximity (within ~2.5 cm) to an electrode. At other stages of development and at lower voltage levels, egg mortality rates were considerably less. Arctic grayling eggs experienced less mortality before and after 70 temperature units post fertilization and chinook salmon eggs experienced less mortality after 131 temperature units post fertilization. Voltage gradient measurements within gravel indicated that gravel redds offer little or no protection to chinook salmon eggs from electroshock other than providing a physical barrier that keeps the eggs at a safe distance from the electrode. The average drop in voltage from the surface of the gravel to a depth of 40 cm was 72%. For comparison, the average drop in V/cm horizontally from the electrodes in the water to 40 cm was 70%. Investigators using electrofishing as a sampling method should be aware of the potential harm to the eggs of targeted fish and also to the eggs of other fish present at the time of sampling when those eggs are close to the electrodes.

Key Words: Arctic grayling, *Thymallus arcticus*, chinook salmon, *Oncorhynchus tshawytscha*, king salmon, least cisco, *Coregonus sardinella*, humpback whitefish, *Coregonus sardinella*, electrofishing, pulsed direct current, mortality, egg mortality, electrofishing injury, voltage gradient.

INTRODUCTION

Electrofishing is an effective method for capturing fish and provides a means to capture fish within river sections that are difficult to sample by other methods (Reynolds 1983; Clark 1985). Electrofishing is a sampling method used to estimate abundance and age composition for Arctic grayling *Thymallus arcticus* in the Chena River, Salcha River, Chatanika River, and Piledriver Slough (Clark 1995; Fleming 1995; Roach 1995); for chinook salmon *Oncorhynchus tshawytscha* in the Chena and Salcha rivers (Evenson 1993; Skaugstad 1993), and for least cisco *Coregonus sardinella* and humpback whitefish *C. pidschian* in the Chatanika River (Fleming 1994). These sampling events occur near the time of spawning for Arctic grayling in Piledriver Slough; chinook salmon in the Chena, Salcha, and Chatanika rivers; and humpback whitefish and least cisco in the Chatanika River.

Most electrofishing research has focused on physiological changes, injury, survival, and growth of fish exposed to electroshock (Hudy 1985; Mesa and Schreck 1989; Sharber and Carothers 1988; Holmes et al. 1990; Roach 1992; Taube 1992; Mitton and McDonald 1994; Dwyer 1995). Recently the effects of electroshock on spawning behavior and survival of eggs have received increased interest. Sorensen (1994) reported that goldfish *Carassius auratus* and brook trout *Salvelinus fontinalis* spawned normally with sexually active conspecifics within 24 h after being electroshocked. Godfrey (1957) and Marriott (1973) conducted some of the first experiments on survival of eggs after electroshock. These along with more recent studies suggest that eggs exposed to electroshock may survive at a lower rate than eggs not exposed to electroshock (Dwyer et al. 1993; Dwyer and Erdahl 1995). Results, however, varied by voltage, distance from the electrode, duration of exposure, and developmental stage of the egg when exposed to electroshock. Furthermore, in some cases, these experiments were carried out at voltages and durations of exposure that are outside the range that fish or eggs would normally be exposed to from typical electrofishing.

Godfrey (1957) reported mortality rates for brook trout eggs exposed to 150 or 550 V DC (0.6 or 1.7 A; voltage gradient within the water was not reported), for 30 to 300 s, 0.3 to 3.7 m from the electrodes at various developmental stages. At the lower voltage, there was no indication that egg mortality resulted from electroshock treatment. At the higher voltage, however, percent mortality of brook trout eggs electroshocked during early development (6-days postfertilization) ranged from 5 to 100%, which varied by duration of electroshock and distance from the electrodes. In contrast, percent mortality of brook trout eggs electroshocked at either the higher or lower voltage, during late-eyed development (44-days postfertilization) ranged from 0 to 10%.

Dwyer et al. (1993) reported similar results for rainbow trout *Oncorhynchus mykiss* eggs exposed to 340 V pulsed DC (0.6 A; 0.9-1.0 V/cm), for 10 s, at various developmental stages. Percent mortality for eggs exposed to electroshock was significantly different from handled controls at early developmental stages (2- to 10-day postfertilization) but was not significantly different from handled controls at late developmental stages (12- to 26-day postfertilization). Mean percent mortality ranged from 20% (18-days postfertilization; handled control = 20%) to 58% (8-days postfertilization; handled control = 30%). In addition, Dwyer et al. (1993) reported that the greatest mortality from mechanical (eggs dropped 15 cm onto a soft plastic bumper) and electrical shock occurred at the same developmental stages. The eggs, however, were more susceptible to mortality from mechanical shock than electrical shock at early developmental stages.

Another study with cutthroat trout *O. clarki* did not indicate a difference in mortality between control eggs and eggs exposed to 30 or 60 Hz, 150 V pulsed DC (~1.4 V/cm) for 10 s at four developmental stages (Dwyer and Erdahl 1995). There was greater mortality, however, for eggs exposed to 30 or 60 Hz, 225 V pulsed DC and continuous DC (~2.2 V/cm). Also, the Coffelt Complex Pulse System (CPS[®]) resulted in greater mortality at 350 and 450 V compared to controls. The investigations of Dwyer et al. (1993) and Dwyer and Erdahl (1995) demonstrate that the effects of electrical shock on fish eggs is dependent on species, voltage waveform, and voltage level.

RESEARCH OBJECTIVES

The goal of the present study was to gain a better understanding of the effects of electrofishing on mortality of eggs for Arctic grayling, chinook salmon, least cisco, and humpback whitefish. In

addition, voltage gradients within gravel were measured to determine if gravel protects chinook salmon eggs from electroshock once deposited within the redd. The research objectives were to examine differences:

- 1) between mean mortality rate of eggs from parents that have been electroshocked and from parents that have not been electroshocked for Arctic grayling, chinook salmon, least cisco, and humpback whitefish; such that an absolute difference of 0.10 in mean mortality rate was detected 90% of the time at an alpha level of 0.05;
- 2) among mean mortality rate of fertilized eggs that have been electroshocked at seven (Arctic grayling) or nine (chinook salmon) different developmental stages; with four (Arctic grayling) or three (chinook salmon) different levels of electricity; and, from parents that have been electroshocked or not electroshocked for Arctic grayling and chinook salmon; such that an absolute difference of 0.10 in mean mortality rate was detected 90% of the time at an alpha level of 0.05; and,
- 3) determine average voltage gradients by depth in 5 cm increments from the surface of the gravel to 40 cm deep and within three different gravel substrates when the electrodes of a standard electrofishing boat were in contact with the gravel.

METHODS

Two experiments were designed to examine the effects of electroshocking on mortality of eggs. For Experiment 1, Arctic grayling, chinook salmon, and least cisco parents were either electroshocked or not electroshocked and egg mortality compared between the two groups (objective one). In addition, the mortality rate of humpback whitefish eggs from an electroshocked group of humpback whitefish was determined. For Experiment 2, Arctic grayling and chinook salmon eggs, from parents electroshocked or not electroshocked, were electroshocked or not electroshocked at various stages of development and egg mortality compared among the groups (objective two). A Kolmogorov-Smirnov two-sample test was used to compare length distributions of fish electroshocked and fish not electroshocked.

For Experiment 1, a comparison of two mean proportions was used to examine the effects of electroshocking fish on egg mortality. The comparison was made between the proportion of eggs that died before the eyed stage of development from parents that were electroshocked (electroshocked group) and the mean proportion of eggs that died before the eyed stage from parents that were not electroshocked (control group). This comparison was made for three resident species: Arctic grayling, chinook salmon, and least cisco. The hypothesis:

$$H_o: \pi_c = \pi_s$$

$$H_a: \pi_c < \pi_s$$

was tested to determine whether there was a difference in mortality rates between the electroshocked and control groups. The test statistic used was:

$$t = \frac{\bar{p}_c - \bar{p}_s}{\sqrt{V[\bar{p}_c] + V[\bar{p}_s]}} \quad (1)$$

where: \bar{p}_c = average mortality for the control group; and,

\bar{p}_s = average mortality for the shocked group.

For Experiment 2, analysis of variance was used to examine the differences among the mean mortality rate of fertilized eggs that were electroshocked at different developmental stages, at different levels of electroshock, and from parents that were electroshocked or not electroshocked for Arctic grayling and chinook salmon.

To transform mean mortality rates into a nearly normal distribution, angular transformation was used on the proportion of dead eggs (Zar 1984):

$$p' = \frac{1}{2} \left[\arcsin \sqrt{\frac{X}{n+1}} + \arcsin \sqrt{\frac{X+1}{n+1}} \right]. \quad (2)$$

Analysis of variance was used on the transformed proportions to examine the effects of electroshocking fertilized fish eggs on egg mortality to the eyed stage of development. This experiment used a randomized block design with a factorial treatment arrangement, all treatments were considered fixed. Probabilities of a type I error (α) of 0.05 or lower were considered significant. The model used was:

$$Y_{ijkl} = \mu + P_i + E_j + D_k + PE_{ij} + PD_{ik} + ED_{jk} + PED_{ijk} + e_{(ijkl)} \quad (3)$$

where: Y_{ijkl} = the angular transformation of the proportion of eggs that died from the l^{th} replicate, shocked at the k^{th} stage of development at the j^{th} level of electroshock from parents of the i^{th} group;

μ = overall mean mortality of all treatments;

P_i = effect of the i^{th} group of parents;

E_j = effect of the j^{th} level of electroshock;

D_k = effect of being shocked at the k^{th} stage of development;

PE_{ij} = effect of the interaction of the i^{th} group of parents and the j^{th} level of electroshock;

PD_{ik} = effect of the interaction of the i^{th} group of parents and being electroshocked at the k^{th} stage of development;

ED_{jk} = effect of the interaction of the j^{th} level of electroshock and being electroshocked at the k^{th} stage of development;

PED_{ijk} = effect of the interaction of the i^{th} group of parents, the j^{th} level of electroshock, and being shocked at the k^{th} stage of development; and,

$e_{(ijkl)}$ = error of the l^{th} replicate associated with the i^{th} group of parents, the j^{th} level of electroshock, and being shocked at the k^{th} stage of development.

In the case where effects were significant and there were no interactions among main effects, Duncan's multiple range test was performed *a posteriori* to determine significance within the effect. Probabilities of a type I error (α) of 0.05 or lower were considered significant.

ARCTIC GRAYLING

In early May 1994, at a weir-trap site at Moose Lake, fishery technicians captured and retained several hundred mature Arctic grayling. On May 9, 1994 investigators selected a sample of 60 female and 60 male Arctic grayling, which were randomly divided into two treatment groups of 30 females and 30 males each. One group was electroshocked and the other was not (control group). Fish were held separately by group and sex until the egg-take, wherein the two groups were spawned separately and the fertilized eggs of the two groups kept separate. The length of each fish was measured to the nearest millimeter and recorded.

For electroshock treatment, Arctic grayling were placed ten at a time in a 1.22 x 1.22 x 1.22 m mesh holding pen and electroshocked for 5 s with a Coffelt[®] Mark-10 variable voltage pulsator (VVP) backpack electroshocker powered by a Honda[®] 350EX gasoline generator. The anode consisted of an aluminum hoop attached to a 1.5 m handle, which was connected to the VVP with a 1.8 m coil cord. A 1.8 m steel rattail served as the cathode. VVP output settings were 200 V, 60 Hz pulsed DC, and 50% duty cycle at 1.3 A. Water conductivity was 340 μ S/cm and water temperature was 5° C. The anode was placed in the center of the holding pen and moved toward the fish. These conditions resulted in the fish being exposed to an average voltage gradient that ranged from 0.14 to 1.40 V/cm as measured with a voltage gradient probe attached to a digital voltmeter. Exact voltage gradients that each fish was exposed to was not known but all fish were stunned in a manner consistent with an actual field situation (i.e., all fish exhibited galvanonarcosis).

For the egg take, the investigators followed a procedure that minimized the natural variability among fish and provided each male in a group an equal opportunity of fertilizing the eggs of each female in that same group. Eggs were removed from females by excising the abdominal wall such that the eggs flowed into a spawning pan. An equal volume of eggs was retained from each of the females from each group (fish electroshocked or fish not electroshocked). From the eggs of each female, a 10 mL sample was taken, added to a pool of eggs for that group (parents electroshocked or not electroshocked), and the excess eggs discarded. The pool of eggs was mixed, 30 equal volumes (10 mL) of eggs were then taken from the pool, and the sperm of one male added to each aliquot separately. These eggs were again mixed and a small amount of water added to activate the sperm, after at least one minute the eggs were washed to remove excess sperm and debris. Eggs of each group were placed into a one liter container for transport back to the hatchery. The containers were clearly marked; "Shocked Fish", or "Control Fish". The two egg containers were placed in an ice cooler and lightly iced to maintain a temperature of approximately 3.5° C. The eggs were flown to Clear Hatchery on the same day as the egg take.

At the hatchery the eggs were enumerated into uniquely numbered fiberglass-screen baskets using a volumetric method without regard to the condition of the eggs. These baskets were placed into Heath trays for incubation. The water flow was 4 to 5 g/m and water temperature was gradually raised from 3.2° to 11.0° C. The eggs were treated with a 15 m, 1:600 solution, formalin drip every other day to control fungal infection.

Experiment 1

Investigators kept eggs from control fish and eggs from shocked fish separate by placement into identifiable fiberglass-screen baskets. A total of six baskets (three replicates from shocked fish and three replicates from control fish) of approximately 600 eggs each were placed into Heath

trays for incubation. Exact counts of eggs within each basket were recorded. One replicate each of shocked and control fish were placed side-by-side in a single Heath tray to minimize front to back variation within a tray. Each replicate was placed in a separate stack of Heath trays.

Experiment 2

Investigators kept eggs from control parents and eggs from electroshocked parents separate by placement into identifiable fiberglass-screen baskets. A total of 168 baskets (three replicates x seven egg-developmental stages x four electroshock levels x two groups of parents) of approximately 100 eggs each were placed in Heath trays for incubation and treatment. Six additional baskets (three from each group) of approximately 100 eggs each were placed in Heath trays for incubation. These additional eggs were used as reference eggs to classify the stage of egg-development at the time of treatments. The baskets were arranged within the Heath trays by a randomized block design: one replicate per stack, four baskets placed within one Heath tray, and the control- and shocked-parent of the same treatment placed side-by-side within a tray. Disturbance to trays was kept to a minimum and all baskets within one tray received treatments on the same day. On the day of each designated stage of development, the predetermined baskets were removed one at a time, placed within the electroshocking tub, electroshocked at the specified level (high, medium, low, or none) for 5 s, and placed back in the Heath tray. Exact counts of eggs within each basket were recorded. On each day of treatment, a sample of eggs from the reference group was retained. The reference eggs were classified by number of days postfertilization, temperature units postfertilization, stage of development, and a physical description.

The electroshocking tub was a one-piece molded rigid plastic tray with metal electrodes at each end that spanned the complete cross section of water in the tank (Roach 1992). Electrical wire connected the electrodes to a Coffelt[®] Model 15 VVP, which delivered a uniform electrical field to the water. Levels of electroshock used for treatments were high (1.30 - 1.50 V/cm), medium (0.80 - 1.00 V/cm), low (0.30 - 0.50 V/cm), or none. The settings on the VVP were adjusted to achieve desired voltage gradients at 60 Hz and 50% duty cycle. To insure consistent voltages within the ranges, power was turned on and average voltage gradients measured with a voltmeter after the initial power surge, but before the eggs were placed in the center of the shocking tub.

CHINOOK SALMON

Investigators captured 29 chinook salmon from the spawning grounds on the upper Salcha River. Eighteen were captured on July 20, 1994 using a 100 x 7 m adult salmon gillnet as a beach seine and 11 on July 21, 1994 by electroshocking using a standard electroshocking boat. Fish captured with the gillnet were marked for later identification. Fish were held separately by sex until the egg-take. On July 24, 1994, five females and five males from each group (electroshocked or not electroshocked) were spawned separately, similar to the method used with the Arctic grayling except that a 500 mL sample of eggs was taken from each fish instead of 10 mL. Only fish with free-flowing eggs were used in the experiment. The eggs were transported to Clear Hatchery the same day as the egg-take and handled there with the same procedures used with Arctic grayling. The length of each fish was measured to the nearest millimeter and recorded.

The electrofishing boat used to capture the electroshocked group of chinook salmon was equipped with a Coffelt[®] Model 15 VVP powered by a 3,500 W single-phase gasoline generator. Anodes consisted of four 15 mm diameter steel cables (1.5 m long) arranged perpendicular to the

long axis of the boat and 2.1 m forward of the bow. The unpainted bottom of the aluminum boat was utilized as the cathode. VVP settings were 250 V, 60-Hz pulsed DC, and 50% duty cycle at 2 A. Water conductivity was 145 $\mu\text{S}/\text{cm}$ and water temperature was 11° C. These conditions resulted in the fish being exposed to an average voltage gradient, as measured with a voltage gradient probe attached to a digital voltmeter, which ranged from 0.07 V/cm in midwater at the midpoint between the outside anodes and the bow of the boat; and 1.24 V/cm in midwater ~2.5 cm from the anodes. Exact voltage gradients that each fish was exposed to were not known but all fish exhibited galvanonarcosis and were easily captured with dip-nets.

Experiment 1

Investigators kept eggs from control fish and eggs from shocked fish separate by placement into identifiable fiberglass-screen baskets. A total of six baskets (three replicates from shocked fish and three replicates from control fish) of approximately 600 eggs each were placed into Heath trays for incubation. Exact counts of eggs within each basket were recorded. One replicate each of shocked and control fish were placed side-by-side in a single Heath tray to minimize front to back variation within a tray. Each replicate was placed in a separate stack of Heath trays.

Experiment 2

Procedures were similar to the Arctic grayling experiment with the exception that 162 baskets (three replicates \times nine egg-developmental stages \times three electroshock levels \times two groups of parents) of approximately 100 eggs each were placed in Heath trays for incubation and treatment instead of 168 baskets. Levels of electroshock used for treatments were high (1.30 - 1.50 V/cm), medium (0.80 - 1.00 V/cm), or none.

Voltage Measurements in Gravel

Voltage gradients within gravel by depth were measured at four different substrate types (silt, gravel and silt, gravel and cobble, and gravel and large cobble) in the Chena River during July 1995. While silt is not likely to be used for spawning, the remaining three substrate types include those that chinook salmon do use for spawning. A standard electrofishing boat as described for Experiment 1 was used to produce voltage gradients within gravel. Settings on the VVP were 60 Hz and 50% duty cycle. Voltage was adjusted at the VVP so that the greatest voltage gradient ~2.5 cm from the electrodes was approximately 1.25 V/cm, which is similar to the maximum voltage used in normal field electroshocking activities (200 to 250 V as measured at the VVP). Conductivity ranged from 135 to 150 $\mu\text{S}/\text{cm}$ and water temperature was 10° C.

The electrofishing boat was positioned such that the electrodes were laying directly on top of the gravel. To measure the greatest voltage gradients at a given point within the gravel, voltage was measured in the plane perpendicular to the electrodes. V/cm was measured at 5-cm increments down to 40 cm within the gravel with a Fluke 29 Series II multimeter and a voltage gradient probe.

Gravel substrate was measured and described by a wet-sieving technique similar to that used by Shirazi et al. (1980). Proportion of silt, gravel, cobble, and large cobble was determined for each site. Gravel was sieved through a series of gravel sieves starting with the largest mesh and progressing to the smallest mesh until all of the gravel was measured. The volume of the total gravel and the gravel with diameter greater than the mesh of each sieve was determined.

LEAST CISCO

Investigators captured 81 mature least cisco from the spawning grounds in the Chatanika River. Thirty-two (12 males and 20 females) were captured on September 26, 1995 using 45.7 x 1.8 m variable mesh gillnets and 49 (19 males and 30 females) on September 27, 1995 by electroshocking using a standard electroshocking boat similar to the boat used to capture chinook salmon. Water conductivity was 138 $\mu\text{S}/\text{cm}$ and water temperature was 6.5° C. Least cisco from each group (electroshocked or not electroshocked) were spawned on the same day as capture, similar to the method used with the Arctic grayling. Only fish with free-flowing eggs were used in the experiment. The length of each fish was measured to the nearest millimeter and recorded. The eggs were transported to Clear Hatchery the same day as the egg-take.

Only Experiment 1 was performed with least cisco eggs. Investigators kept eggs from control fish and eggs from shocked fish separate by placement into identifiable fiberglass-screen baskets. A total of six baskets (three replicates from shocked fish and three replicates from control fish) of approximately 600 eggs each were placed into Heath trays for incubation. Exact counts of eggs within each basket were recorded. One replicate each of shocked and control fish were placed side-by-side in a single Heath tray to minimize front to back variation within a tray.

HUMPBACK WHITEFISH

Investigators captured 46 mature humpback whitefish (23 males and 23 females) on the spawning grounds on the Chatanika River, on September 27, 1995 by electroshocking using a standard electroshocking boat similar to the boat used to capture chinook salmon. Water conductivity was 138 $\mu\text{S}/\text{cm}$ and water temperature was 6.5° C. These fish were spawned on the same day as capture, similar to the method used with the Arctic grayling. Only fish with free-flowing eggs were used in the experiment. The length of each fish was measured to the nearest millimeter and recorded. The eggs were transported to Clear Hatchery the same day as the egg-take.

Investigators were unsuccessful at capturing a control group of humpback whitefish (i.e., humpback whitefish by methods other than electrofishing). Humpback whitefish eggs from parents that were electroshocked were handled similar to least cisco eggs. Three baskets of approximately 600 eggs each were placed into Heath trays for incubation. Exact counts of eggs within each basket were recorded.

RESULTS

For Arctic grayling, egg mortality rates were low and differences in mean mortality rates between controls and electroshocked groups were less than 10 percentage points. For chinook salmon, egg mortality rates were high and differences in mean mortality rates between controls and electroshocked groups were, in most cases, greater than 10 percentage points. For least cisco, egg mortality rates were high, but differences in mean mortality rates between controls and electroshocked groups were less than 10 percentage points. For humpback whitefish, egg mortality was high. Although not comparable to a control, the mean mortality rate for humpback whitefish eggs was less than the mean mortality rate for either the control or treated eggs from least cisco parents. The eggs of least cisco and humpback whitefish are similar in size, rate of development, and ontogeny.

Table 1.-Mean percent mortalities and standard errors for eggs from parents electroshocked and from parents not electroshocked by species.

| Species | Egg Mortality | | | |
|--------------------|---------------------|-------|-----------------|-------|
| | Parents Not Shocked | | Parents Shocked | |
| | Percent | SE | Percent | SE |
| Arctic Grayling | 0.020 | 0.003 | 0.036 | 0.004 |
| Chinook Salmon | 0.012 | 0.002 | 0.200 | 0.009 |
| Least cisco | 0.520 | 0.012 | 0.568 | 0.011 |
| Humpback whitefish | - | - | 0.414 | 0.011 |

ARCTIC GRAYLING

Lengths of the Arctic grayling used in the experiments ranged from 263 to 362 mm FL for females and 254 to 378 mm FL for males. Mean lengths for Arctic grayling electroshocked were 323 mm FL for females and 316 mm FL for males. Mean lengths for Arctic grayling not electroshocked were 318 mm FL for females and 313 mm FL for males. There were no significant differences between the lengths of Arctic grayling electroshocked and Arctic grayling not electroshocked (males, $D = 0.17$, $P = 0.79$; females, $D = 0.20$, $P = 0.59$).

Experiment 1

Mean egg mortality rate was 0.02 for Arctic grayling eggs from parents that were not electroshocked compared to 0.04 for Arctic grayling eggs from parents that were electroshocked (Table 1; $t = -2.96$, 4 d.f., $P = 0.02$).

Experiment 2

Classification of Arctic grayling egg developmental stages from reference eggs at the time of electroshock treatment ranged from the morular stage through final stages of epiboly (Table 2).

Analysis of variance indicated that all main effects were significant with no interactions (Table 3). The greatest mean mortality rates occurred at 70-TU postfertilization. At this stage of development mean mortality rate was 0.12 (SE = 0.05) for Arctic grayling eggs electroshocked at the high level (1.30 to 1.50 V/cm) from parents that were electroshocked before spawning; compared to 0.07 (SE = 0.04) for eggs electroshocked at the high level from parents not electroshocked; and compared to 0.04 (SE = 0.02) for eggs not electroshocked from parents that were also not electroshocked (Figure 1). The least mean mortality rates occurred at 8-TU postfertilization. At this stage of development mean mortality rate was 0.02 (SE = 0.01) for Arctic grayling eggs not electroshocked from parents not electroshocked; compared to 0.02 (SE = 0.02) for eggs not electroshocked from parents electroshocked; and compared to 0.07 (SE = 0.04) for eggs electroshocked at the high level from parents electroshocked (Figure 1).

The results of Duncan's multiple range comparison tests indicated significant differences between mean mortality rates for:

- 1) Arctic grayling eggs electroshocked from parents electroshocked (0.06) compared to eggs electroshocked from parents not electroshocked (0.04);

Table 2.-Brief description of Arctic grayling eggs at the time of treatments (day postfertilization), number of temperature units postfertilization (TU), and developmental stages (stage).

| Day | TU | Stage | Brief Description |
|-----|-----|-----------|---|
| 2 | 8 | Morular | Elevated blastodisc |
| 4 | 22 | Gastrular | Flattened podium |
| 6 | 38 | Epiboly | Yolk plug large; neural plate visible |
| 8 | 54 | Epiboly | Optic vesicles visible; tail slightly raised |
| 10 | 70 | Epiboly | Tail raised < 1/2; brain lobes differentiated |
| 12 | 86 | Epiboly | Tail raised > 1/2; head slightly raised; faint eye pigmentation |
| 14 | 102 | Epiboly | Dark eye pigmentation; brain obviously swollen |

- 2) all combinations of eggs electroshocked by level of electroshock; high (0.07), medium (0.06), low (0.04), and none (0.04);
- 3) eggs electroshocked at 8 TU (0.04) compared to all other times; 22 TU (0.05), 38 TU (0.05), 54 TU (0.06), 70 TU (0.07), 86 TU (0.06), and 102 TU (0.05);
- 4) eggs electroshocked at 70 TU compared to all other times;
- 5) eggs electroshocked at 22 TU compared to 8 TU, 70 TU, and 86 TU; and,
- 6) eggs electroshocked at 86 TU compared to 8 TU, 22 TU, and 70 TU.

CHINOOK SALMON

Lengths of the chinook salmon used in the experiments ranged from 795 to 975 mm FL for females and 625 to 825 mm FL for males. Mean lengths for chinook salmon electroshocked were 890 mm FL for females and 724 mm FL for males. Mean lengths for chinook salmon not electroshocked were 883 mm FL for females and 730 mm FL for males. There were no significant differences between the lengths of chinook salmon electroshocked and chinook salmon not electroshocked (males, $D = 0.40$, $P = 0.82$; females, $D = 0.20$, $P = 0.99$).

Experiment 1

A mean egg mortality rate of 0.20 for chinook salmon eggs from parents that were electroshocked was significantly greater than the mean egg mortality rate of 0.01 for chinook salmon eggs from parents that were not electroshocked ($t = -20.03$, 4 d.f., $P < 0.01$; Table 1).

Experiment 2

Classification of chinook salmon egg developmental stages from reference eggs at the time of electroshock treatment ranged from the morular stage through final stages of epiboly (Table 4).

Analysis of variance indicated that all main effects were significant with significant interactions between parent and developmental stage; and between level of electroshock and developmental stage (Table 5). The greatest mean mortality rate (1.00) occurred for chinook salmon eggs electroshocked at the high level, at 98-TU postfertilization, and from parents that were

Table 3.-Source of effects, degrees of freedom (df), sum of squares, mean of squares, F-values, and P-values for ANOVA of Arctic grayling Experiment 2. Sources of effects were parents (P), level of electroshock (E), and egg developmental stage (D). Significant P-values highlighted in bold type.

| Source | df | Sum of Squares | Mean of Squares | F value | P value |
|--------|----|----------------|-----------------|---------|---------------|
| P | 1 | 0.0783 | 0.0783 | 87.93 | 0.0001 |
| E | 3 | 0.1460 | 0.0487 | 54.64 | 0.0001 |
| P*E | 3 | 0.0039 | 0.0013 | 1.45 | 0.2330 |
| D | 6 | 0.0661 | 0.0110 | 12.36 | 0.0001 |
| P*D | 6 | 0.0023 | 0.0004 | 0.43 | 0.8594 |
| E*D | 18 | 0.0149 | 0.0008 | 0.93 | 0.5472 |
| P*E*D | 18 | 0.0088 | 0.0005 | 0.55 | 0.9271 |

electroshocked before spawning; compared to eggs not electroshocked, handled at the same time, but from parents not electroshocked (0.18; SE = 0.04); and compared to eggs handled at the same time, from parents not electroshocked, but eggs not electroshocked (0.03; SE = 0.02; Figure 2). The least mean mortality rate (0.01; SE = 0.01) occurred for chinook salmon eggs not electroshocked, handled at 240-TU postfertilization, and from parents not electroshocked; compared to eggs not electroshocked, handled at the same time, but from parents electroshocked (0.17; SE = 0.06); and compared to eggs from parents electroshocked, handled at the same time, but electroshocked at the high level (0.31; SE = 0.07; Figure 2).

Significant interactions between the main effects of the chinook salmon Experiment 2 precluded performing Duncan's multiple range comparison tests. This interaction was significant because the magnitude of the difference between the shocked and not shocked parents and levels of electroshock depended on the stage of development.

Voltage Measurements in Gravel

The substrate of the four sites varied from mostly silt to gravel and large cobble (Figure 3). Within the first 10 cm of gravel, the rate of decrease in V/cm was greatest for sites with the largest gravel sizes (Figure 4). The average drop in voltage from the surface of the gravel to a depth of 40 cm was 72% (1.19 V/cm to 0.33 V/cm). For comparison, the average drop in V/cm horizontally from the electrodes in the water to 40 cm was 70% (1.24 V/cm to 0.37 V/cm).

LEAST CISCO

Lengths of least cisco used in the experiments ranged from 297 to 410 mm FL for females and 294 to 365 mm FL for males. Mean lengths for least cisco electroshocked were 355 mm FL for females and 334 mm FL for males. Mean lengths for least cisco not electroshocked were 356 mm FL for females and 336 mm FL for males. There were no significant differences between the

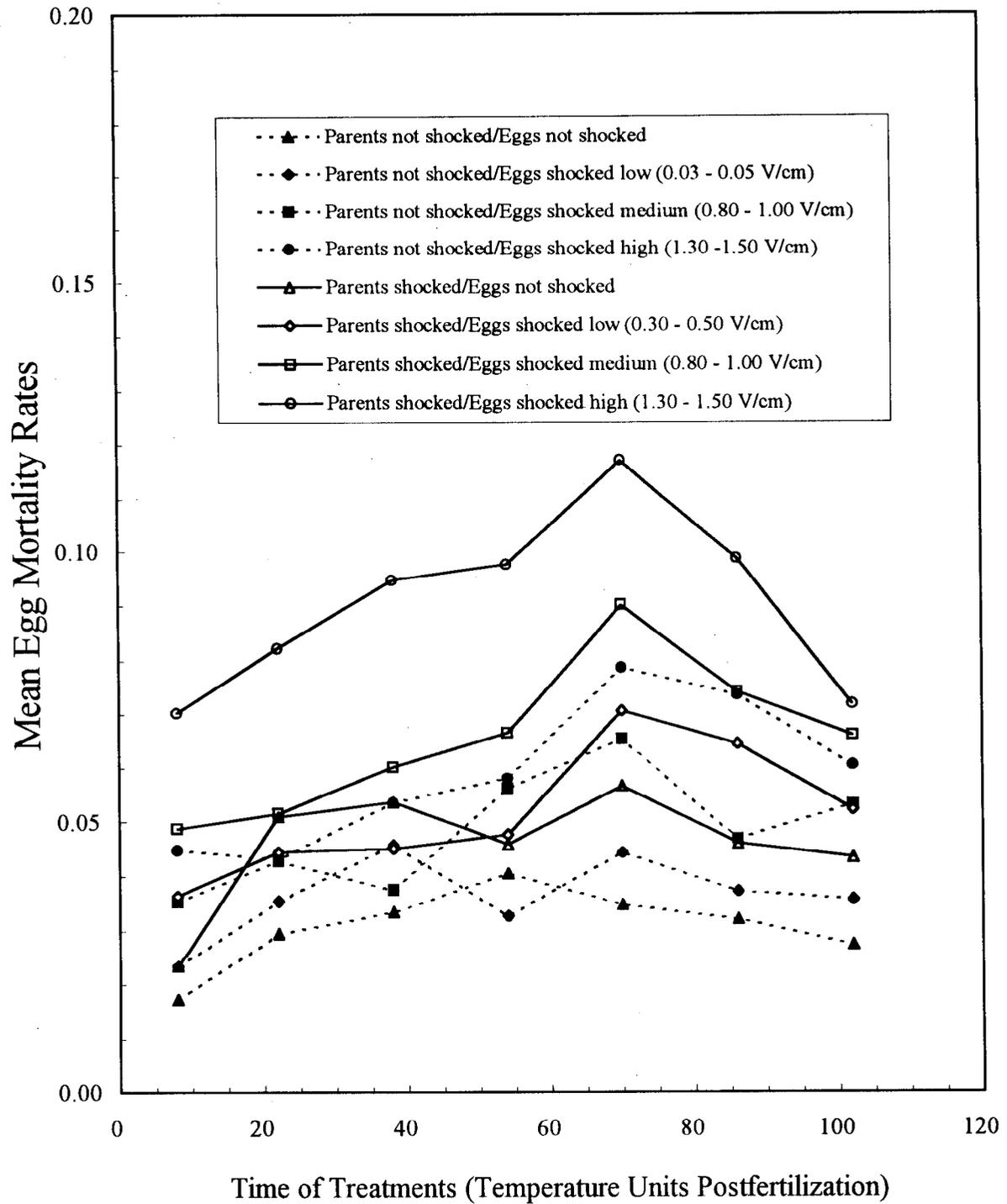


Figure 1.-Mean mortality rates for electroshocked Arctic grayling eggs by time of treatments (temperature units postfertilization), parents (not electroshocked or electroshocked), and electroshock levels (none, low, medium, or high).

Table 4.-Brief description of chinook salmon eggs at the time of treatments (day postfertilization), number of temperature units postfertilization (TU), and developmental stages (stage).

| Day | TU | Stage | Brief Description |
|-----|-----|----------------|--|
| 3 | 33 | Morular | Elevated blastodisc |
| 6 | 65 | Gastrular | Distinct terminal node |
| 9 | 98 | Late Gastrular | Germ ring at the equator |
| 12 | 131 | Epiboly | Yolk plug still present but diminishing |
| 14 | 153 | Epiboly | Optic vesicles visible; brain lobes differentiated |
| 16 | 175 | Epiboly | Tail slightly raised |
| 18 | 197 | Epiboly | Head slightly raised, faint eye pigmentation |
| 20 | 219 | Epiboly | Tail raised > 1/2, dark eye pigmentation |
| 22 | 240 | Epiboly | Pectoral fins readily visible, head raised |

lengths of least cisco electroshocked and least cisco not electroshocked (males, $D = 0.29$, $P = 0.54$; females, $D = 0.17$, $P = 0.89$).

A mean egg mortality rate of 0.57 for least cisco eggs from parents that were electroshocked was significantly greater than the mean egg mortality rate of 0.52 for least cisco eggs from parents that were not electroshocked ($t = -3.203$, 4 d.f., $P = 0.02$; Table 1).

HUMPBACK WHITEFISH

Lengths of humpback whitefish used in the experiments ranged from 369 to 470 mm FL for females and 376 to 480 mm FL for males.

Table 5.-Source of effects, degrees of freedom (df), sum of squares, mean of squares, F-values, and P-values for ANOVA of chinook salmon Experiment 2. Sources of effects were: parents (P), level of electroshock (E), and egg developmental stage (D). Significant P-values highlighted in bold type.

| Source | df | Sum of Squares | Mean of Squares | F value | P value |
|--------|----|----------------|-----------------|---------|---------------|
| P | 1 | 3.4210 | 3.4210 | 293.73 | 0.0001 |
| E | 2 | 8.8866 | 4.4433 | 381.51 | 0.0001 |
| P*E | 2 | 0.0459 | 0.0230 | 1.97 | 0.1443 |
| D | 8 | 9.2237 | 1.1530 | 99.00 | 0.0001 |
| P*D | 8 | 0.5237 | 0.0655 | 5.62 | 0.0001 |
| E*D | 16 | 7.3091 | 0.4568 | 39.22 | 0.0001 |
| P*E*D | 16 | 0.2746 | 0.0172 | 1.47 | 0.1229 |

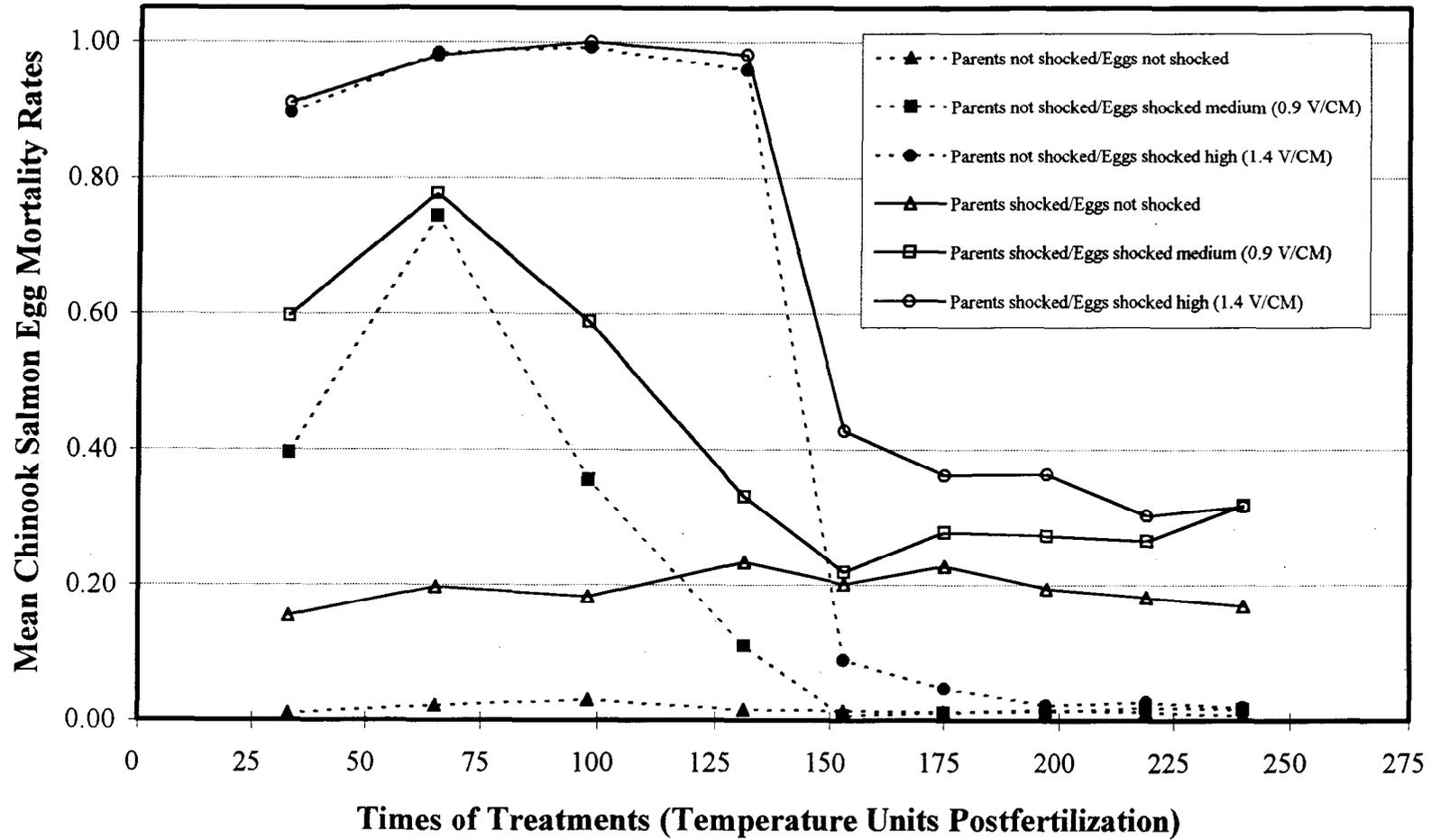


Figure 2.-Mean mortality rates for electroshocked chinook salmon eggs by time of treatment (temperature units postfertilization), parent (not electroshocked or electroshocked), and electroshock level (none, medium, or high).

Egg mortality rate was 0.41 for humpback whitefish eggs from parents that were electroshocked (Table 1). There was not a control group for humpback whitefish eggs from parents that were not electroshocked.

DISCUSSION

The results of the Arctic grayling, chinook salmon, least cisco, and humpback whitefish egg mortality experiments support the hypothesis that mortality of fish eggs is influenced by electroshock. Furthermore, this study suggested that mortality of fish eggs from electroshock is related to the species, magnitude of the electroshock, the stage of egg development, and whether the parents were electroshocked or not. The sample sizes used in these experiments enabled the detection of small differences in mortality between the control groups and the electroshocked groups and in some cases such as for Arctic grayling and least cisco the differences were negligible.

Fishery biologists routinely accept harm to individual fish as a cost of gaining valuable information on a population of fish. Investigators using electrofishing as a sampling method, however, should be aware of the potential harm to the eggs of the targeted fish and also to the eggs of other fish present at the time of sampling when those eggs are in close proximity to the electrodes (within 2.5 cm). The difference in mortality between the control and electroshocked groups should be considered a cost of obtaining information, which should be compared with the costs of not sampling and the costs of sampling by other methods. Benefits of electrofishing, other than sampling efficiency, should also be considered. For example, Mitton and McDonald (1994) reported that rainbow trout experienced reduced adverse response to air exposure when handled under the mild narcosis following exposure to pulsed DC.

Studies that propose the use of electrofishing as a sampling method should include a synopsis of

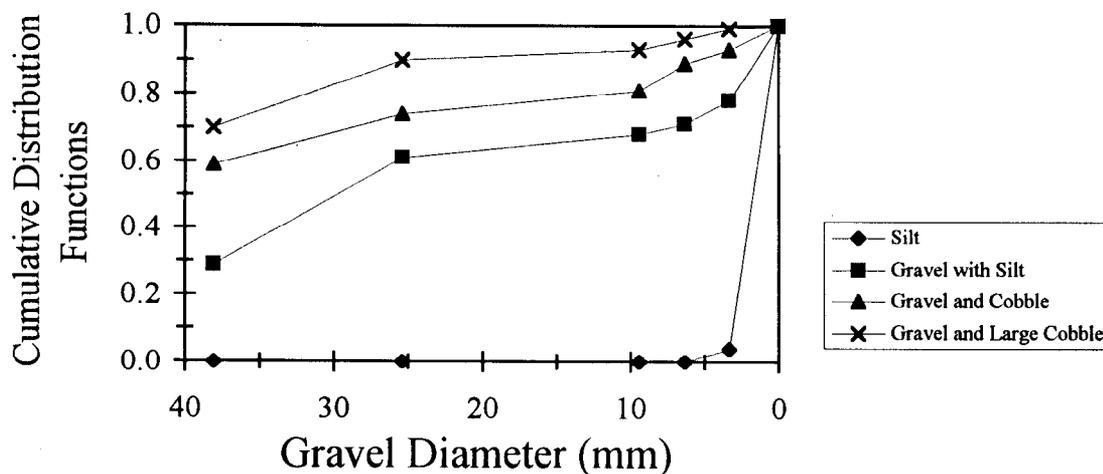


Figure 3.-Cumulative distribution functions of gravel diameters at four sites within the Chena River where voltage gradients were measured.

the costs and benefits in terms of harm to fish involved. Knowledge of the harm to individual eggs from electrofishing can be equated to costs to the population (Schill and Beland 1995). Along with mortality rates, probabilities of being exposed to lethal levels of electroshock should be considered when determining the costs to the population. For most sampling schemes, only a proportion of the population is exposed to the lethal levels of electroshock. For some sampling strategies or for populations condensed within small areas, the probability of exposing a fish to electroshock or the probability of passing over a redd increases and should be evaluated accordingly. Investigators should be aware that, as capture probability increases, egg mortality as a function of population size may also increase.

For both Arctic grayling experiments, egg mortality rates were low and differences in mortality rates between controls and electroshocked groups, in all cases, were less than 10 percentage points. For practical purposes the differences in mortality rates were negligible and should not preclude using electroshock to capture Arctic grayling, as long as the intensity of electroshock is similar to what was used during this study. The greatest effect, however, was on Arctic grayling eggs electroshocked during middle to late epiboly (50 - 90 TU postfertilization; seven to 13 days after spawning at 7° C average water temperature; middle to late May for most interior Alaska rivers). The effects of electroshocking on the mortality of Arctic grayling eggs, even though minimal, may be minimized further by not electroshocking during this time of increased egg sensitivity.

For chinook salmon Experiment 1, the difference in mortality rates between eggs from parents that were electroshocked (0.20) compared to eggs from parents not electroshocked (0.01) was greater than 10 percentage points. The difference in egg mortality rate (0.19) is a maximum mortality rate for the eggs of prespawning chinook salmon exposed to electroshocking. Given a probability of less than 0.10 that a particular fish from a stock is exposed to the level of

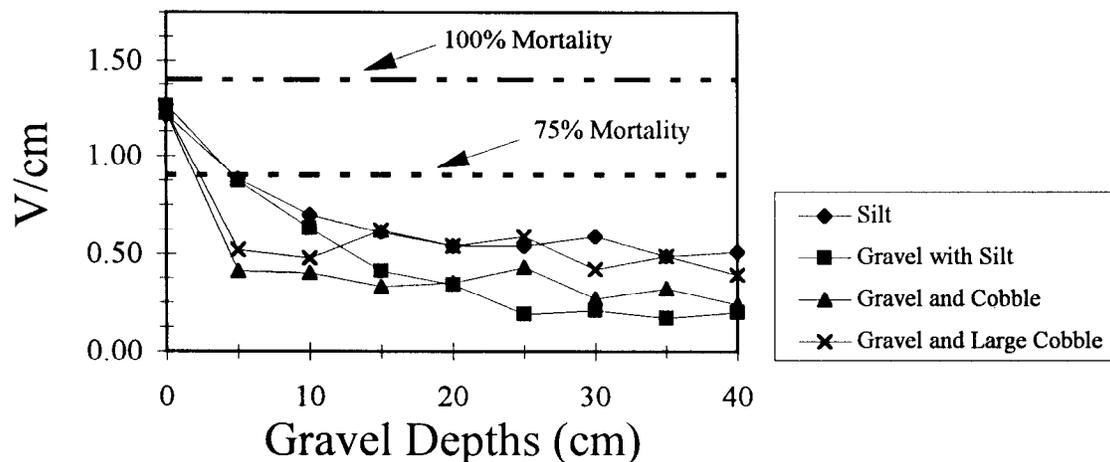


Figure 4.-V/cm measured by gravel depth and substrate type with the electrode of a standard electroshocking boat laying on top of the gravel.

electroshock that would enable it to be captured during a sampling event (Evenson 1993; Skaugstad 1993), there is not a need for a complete moratorium on electroshocking in waters with prespawning chinook salmon. The cost in egg loss can be equated to the loss of females from the stock. For example, given a stock of 10,000 chinook salmon, a male to female ratio of 60:40, and 0.10 probability of capture, electrofishing at present levels and at a time that 75% of the stock has spawned (late July in the Salcha and Chena rivers) is equivalent to removing 19 prespawning females or less than 1% of the female population.

For chinook salmon Experiment 2, egg mortality was high and differences in mortality rates between controls and electroshocked groups were, in most cases, also greater than 10 percentage points and sometimes much greater. Chinook salmon eggs exposed to electroshock at the high level (1.30 - 1.50 V/cm; similar to the voltage gradient within 2.5 cm of the electrodes under normal electroshocking conditions) experienced a mortality rate that ranged from 0.91 to 1.000 during the early to middle stages of development (33 - 131 TU postfertilization; three to 13 days after spawning at 10° C average water temperature; late July in the Salcha and Chena rivers).

The measured voltage gradients within gravel indicated that gravel offered little protection to eggs from electroshock other than as a physical barrier that may keep the eggs at a safe distance from the electrode. In most cases, chinook salmon eggs are buried deeper than 10 cm within the redd. Vronskiy (1972) reported that chinook salmon eggs were buried within the redd at depths between 10 to 80 cm. Briggs (1953) reported 28 cm as the average depth and 20 to 36 cm as the range of depth that chinook salmon buried eggs in two California streams. At these depths, chinook salmon eggs will experience less mortality than reported in this study. Chinook salmon eggs within redds would be exposed to less than the medium level of electroshock used in the experiments of this study (0.90 V/cm). At this level of electroshock and during critical stages of development there was a maximum difference in mortality rates of 76 percentage points when compared to controls (Figure 2). Of course, the deeper the eggs are buried the lower the voltage level. However, using 0.76 as the egg mortality rate from electroshocking directly over a redd as a maximum, mortality can be equated to loss of females from the stock. For example, given a stock of 10,000 chinook salmon, a male to female ratio of 60:40, a probability of 0.14 that a egg is at the critical stage of development, and a probability of 0.10 that the electrodes will pass directly over a redd, electrofishing at present levels and at a time that 75% of the stock has spawned is equivalent to removing a maximum of 32 prespawning females or less than 1% of the female population.

Considering both types of mortality, the cost in egg mortality of using electrofishing to sample a population of 10,000 chinook salmon under the conditions mentioned above is equivalent to removing 51 prespawning females or 1.3% of the female population.

Least cisco egg mortality rates were high but differences in average mortality rate between control eggs (0.52) and eggs from parents exposed to electroshock (0.57) was less than 10 percentage points. For practical purposes the difference in mortality rate was small and should not preclude using electroshock to capture least cisco, as long as the intensity of electroshock is similar to what was used during this study. The cost in egg loss can be equated to the number of females not allowed to spawn. For example, given a stock of 30,000 prespawning least cisco, a male to female ratio of 50:50, and 0.10 probability of capture (Fleming 1994), electrofishing at present levels is equivalent to not allowing 75 females the opportunity to spawn or less than 1% of the female population.

The humpback whitefish experiment was not a controlled experiment. Eggs were only taken from humpback whitefish that were captured by electrofishing. Humpback whitefish and least cisco eggs are similar in size, rate of development, and ontogeny, however, and the effects of electroshocking the parents on egg mortality rate are probably similar (0.05). As with the least cisco eggs, a large part of the humpback whitefish egg mortality rate may be attributed to handling of the eggs. Assuming, however, that the effect on mortality rate is greater than (0.05), the minimum cost in egg loss can be expressed as numbers of females not allowed to spawn. For example, given a stock of 14,000 prespawning humpback whitefish, a male to female ratio of 50:50, and 0.20 probability of capture (Fleming 1994), electrofishing at present levels is equivalent to not allowing a minimum of 70 females the opportunity to spawn or 1% of the female population.

Overall, the results of this study demonstrate the potential harm of electrofishing to individual fish eggs, most notably to chinook salmon eggs and less to Arctic grayling and least cisco eggs. However, using realistic estimates of capture probabilities along with the mortality rates determined by this study, project biologists who want to make a decision about using electrofishing as a sampling tool can and should estimate the costs to the population before making that decision. Several examples of this process were given and even though the information used (capture probabilities or probabilities of lethal exposure, male to female ratios, percent of stock that already spawned, and percent of eggs that will be at a critical stage of development at a given time) came from recent studies, each new study will be different and costs to the population should be calculated for each.

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