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MEDICINE, HEALTH & SAFETY 13 FISH
DOE ARCHIVES

PROJECT CHRONOLOGY CHART AND SUMMARY*

of the

Applied Fisheries Laboratory

University of Washington
Seattle, Washington

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* Revised March 11, 1948

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DOE ARCHIVES

PROJECT CHRONOLOGY CHART AND SUMMARY*

of the

Applied Fisheries Laboratory

University of Washington
Seattle, Washington

PROJECT I. Basic Studies of the Effect of X-rays upon Fish in Various Stages of Development.

SECTION I.

Chinook salmon adults, females and males, from Green River. In October 1943 a total of 21 fish were given whole body doses of 25, 50 and 100 r. 11 others used as controls.

Sexual Maturity

Fish were mature when irradiated.

Spawning

Fish were spawned October 31 to November 5, 1943.

Generations

A total of 55,084 eggs were obtained. Their development is outlined in Section II.

Termination

The irradiated spawners died between October 31 and November 11, 1943. Male deaths were natural but females were sacrificed for spawning. See Section II for consideration of offspring.

SECTION II.

Chinook salmon eggs from Section I, totalling 55,084. Observations were made during periods from: Spawning to eyed egg stage, October 31 to November 26, 1943; eyed egg stage to hatching, November 23 to December 24, 1943; hatching to feeding, December 20, 1943 to February 14, 1944; and feeding to liberation February 14 to May 11, 1944.

* Revised March 11, 1948.

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Feeding to Sexual Maturity

A total of 4835 fingerling (from parents exposed to 100 r) were marked between April 29 and May 8, 1944 by the removal of the adipose and left ventral fins and released in the Samish River on May 11, 1944. A total of 4844 fingerling from control parents were marked by the removal of the adipose and right ventral fins and released in the Samish River on May 12, 1944. Returns are expected in the fall of 1946, 1947 and 1948.

Spawning

Returns from the sea: Summer and Fall 1946, the commercial fishery yielded two chinook salmon bearing the marks assigned to and undoubtedly belonging to this experiment. Besides these two salt-water recoveries 23 marked adults returned to the Samish River in the fall of 1946. Of these, 12 including only 2 females, were from control lots and 11, also including only 2 females were the offspring of parents irradiated with 100 r.

Summer and Fall 1947, the 38 irradiated- and control-stock chinook salmon adults retaken, produced 15 egg lots. The spawn from 13 of the females was apparently ripe and normal, while the spawn from two females (lot 13 and 14) appeared to be immature or "green". Further recoveries at Samish are expected in 1948.

Generations

Progeny of 1946 spawners. The eggs of each female taken in fall of 1946 were fertilized by correspondingly treated males, giving 4 groups of eggs. The progeny from the two 100 r - treated females suffered markedly greater mortality than the two control groups.

Progeny of 1947 spawners. The difference in egg mortality of control and irradiated stock up to January 1948 was significant, being greater for the latter group.

Reports

In August 1945 a report of the statistical analysis of the mortalities up to the time of planting was sent to the Rochester Group for criticism. It was returned with helpful suggestions in December 1945.

Progress in Sections I and II was summarized through May 1947 in Report No. UWFL-6, entitled "Preliminary report concerning X-ray effects upon chinook salmon (Oncorhynchus tschawytscha Walbaum) observed through more than one generation". This report was submitted August 1, 1947. Appendices will be written as marked fish return from the sea.

SECTION III.

Mature sockeye salmon adults, females and males, from Cultus Lake, B. C. were treated in November 1943; 24 fish were irradiated with 25, 50 and 100 r and there were 10 controls.

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Spawning

Fish were spawned November 28, 1943 to December 5, 1943.

Termination

The deaths for the males were natural, the females were sacrificed when spawned. The eggs, totalling 46,722, and their development are considered in Section IV of this chart.

SECTION IV.

Sockeye salmon eggs from Section III. Eggs numbering 46,722, spawned on November 28, 1943, eyed on January 4, 1944, hatched between February 17 and February 28, and started to feed April 10. 23,655 of the 25 and 50 r groups were liberated as fingerling on May 31, 1944. The remaining fingerling of the control and the 100 r groups were marked from October 19 to 26, 1944 and planted at Cultus Lake, B. C., on February 20 and 22, 1945. Liberated controls numbered 4139 and bore the mark adipose and right ventral fin. Liberated 100r - 100r fingerling numbered 2404 and had the adipose and left ventral fins removed.

Returns were expected in 1946, 1947, and 1948.

Reports

In December 1945 the statistical analyses of the mortalities of Sections III and IV were completed, but the report was not submitted because it seemed advisable to wait until possible 1946 and 1947 returns from Cultus Lake had been recovered. It was then decided to revise the analysis in the light of the comments of the Rochester Group on the statistical analysis of Section I and II.

In the fall of 1946 and 1947 returning sockeye salmon were carefully examined for marked fins at Cultus Lake, but no marked fish were observed.

Termination

If no marked fish return in 1948, as now appears probable, results will be restricted to mortality patterns of the young fish.

SECTION V.

Steelhead trout adults, females and males, from Columbia River.

Treatment

On December 1 to 6, 1943 five fish were treated (two each to 250 and 500 r and one to 1,000 r) and ten retained as controls.

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Termination

These fish were caught November 24, 1943 at Rock Island, and by the time they were irradiated they were so weak with copepod (Salmincola edwardsii) infection and had developed such extensive fungus infection that they died between December 5 and 15, 1943. The problem contributes little of use and the section was cancelled.

SECTION VI.

Chinook salmon eyed eggs from Green River.

Treatment

From December 19 to 21, 1943 a total of 11,960 eggs divided into 8 lots containing 931 - 974 eggs each were X-rayed. Two lots each were exposed to 25, 50 and 100 r of X-ray and two were used as controls. Hatching, January 22, 1944. Absorption of yolk, March 15, 1944. Liberation, June 2, 1944.

Termination

This section was expanded by the addition of Section VII.

SECTION VII.

Chinook salmon eyed eggs from Green River.

Treatment

Between January 25, and 28, 1944 a total of 4,065 eggs in 13 lots averaging 315 eggs per lot were X-rayed. Exposures were 250, 500, 1,000, 2,500, 5,000 and 10,000 r, with 2 lots reserved for controls. Each exposure was administered to two lots except the 10,000 r exposure which was given to only one lot.

Treatment to hatching, January 25 to February 19, 1944. Hatching to absorption of yolk, February 14 to April 10. Feeding to liberation, April 6 to June 2, 1944.

Termination

The fish remaining after sampling were liberated without being marked.

Reports

A report combining the results of Sections VI and VII, numbered UWFL-2 and entitled "Studies of the Effects of Roentgen Rays on the Growth and Development of the Embryos and Larvae of the Chinook Salmon (Oncorhynchus tschawytscha)", was submitted in the spring of 1946. This report included statistical analyses of the effects upon mortality

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weight, length, cell counts in hemopoietic tissues, kidney, etc., with a qualitative analyses on other tissues.

The work completed in Sections VI and VII was summarized in a final report, UWFL-8, entitled "The Effects of Roentgen Rays on the Embryos and Larvae of the Chinook Salmon." This report included statistical observations on gonads and the tissues that were not considered in the first report. Report No. UWFL-8 was forwarded to the Atomic Energy Commission in December 1947 for clearance and anticipated publication.

SECTION VIII.

January 18 to 20, 1944 male silver salmon were exposed to 25, 50, 100, 250, 500, and 1,000 r.

Treatment

One fish was used for each exposure and two were retained as controls.

Termination

Fish died between January 20 and February 3, 1944. Deaths were natural. Animals were used for study of effect on skin and certain other tissues, such as intestine.

Reports

This section was reported on November 14, 1946 under the title, "Histological Effect of X-rays on Adult Male Silver Salmon, Oncorhynchus kisutch (Walbaum)". Report No. UWFL-4. The report consists chiefly of photographs of histological sections of tissues. Effects of irradiation could not be detected.

SECTION IX.

In February 1944 12 rainbow trout adult fish from the University of Washington were treated to 500, 1,000, and 2,500 r.

Treatment

There were 4 fish for each exposure and numerous controls.

Spawning

Between February 12 and March 18, 1944 these fish were spawned.

Generation

The eggs developed to eyed stage between February 12 and April 8, 1944, and developed from eyed stage to hatching between March 20 and May 10, 1944.

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Termination

Death of adult fish occurred March 17 to May 5, 1944 because of infection with fungus and copepods brought in by the steelhead trout in Section 5. Section discarded.

SECTION X.

A total of 5,400 chinook salmon fingerling from Green River were treated May 22 to 25, 1944.

Treatment

Dosage was 100, 250, 500, 750, 1,000, 1,250, 2,500 and 5,000 r for each of eight lots with one lot retained as control. There were 600 fish in each lot.

Termination

The surviving fingerlings were liberated August 19, 1944 and the preserved material used for histological study.

Reports

On October 29, 1946 a report, numbered UWFL-3, entitled, "Section 10 of Series of Experiments Involving the Effect of X-ray on Fishes: Fingerling Chinook Salmon (Oncorhynchus tschawytscha Walbaum)", was submitted. This report included statistical analyses of the effects upon mortality, weight, length, cell counts in circulating blood, and hemopoietic cell counts in the kidney. Material was preserved at the time of the experiment for further histological studies but present plans do not include its utilization.

Publication

A declassified condensation of this report numbered UWFL-3a entitled "The Effects of X-ray on Mortality, Weight, Length, and Counts of Erythrocytes and Hematopoietic Cells in Fingerling Chinook Salmon, Oncorhynchus tschawytscha Walbaum", has been submitted to Growth for publication.

SECTION XI.

Rainbow trout yearlings, females and males, from University of Washington stock.

Treatment

The fish were exposed to 50, 100, 500, 750, 1,000, 1,500 and 2,500 r. A total of 128 yearlings (nearly two years old) were irradiated in January, 1945. 20 fish were used in each group except the 1,500 r lot which had 16 and the 2,500 r lot which had 12. An additional 20 fish were used as controls.

Feeding to Sexual Maturity

Studies on mortality, growth, and pathological conditions are to be reported upon at a later time.

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Spawning

The trout when spawned for the first time in the spring of 1945 produced the eggs used for Section XI-A. These eggs were reared and the adults spawned to produce eggs of Section XI-B which became mature and were spawned in the spring of 1947, producing the F_2 generation of the original treated group.

The trout of Section XI were spawned for the second time in the spring of 1946 and the offspring (Section XI-C) were saved.

Termination

The parent fish were planted in Green Lake in the summer of 1946.

SECTION XI-A

Rainbow eggs from Section XI.

Feeding to Sexual Maturity

This first spawn (Spring 1945) totaled 115,401 eggs. The eggs were secured from 67 of the 71 females and divided into 134 lots. The successive stages of development of eggs, fry, fingerling, and adults were studied with data on mortality, rate of growth, malformations, etc. obtained.

Generations

These trout spawned in the spring of 1947 and the reproduction is summarized in Section XI-B.

Termination

The adult fish (F_1 of Section XI) were released in June 1947 as further study on them was not thought to be advisable.

SECTION XI-B

These rainbow eggs from Section XI-A are the second generation offspring of the adult fish X-rayed and spawned in 1945.

Data has been collected on the development of these eggs and will be summarized in a forthcoming report.

Termination

The lack of space made it necessary to reduce these offspring (F_2) periodically. However, in each irradiation group a certain number were retained and are being observed for possible F_3 generation studies.

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SECTION XI-C.

The fish in this section represent offspring of Section XI. The Section XI irradiated fish were spawned for the second time in the spring of 1946. Mortality and abnormality data was collected from time of spawning through eyed egg, hatching and feeding stage.

Termination

The fry were planted by State Game Department in the summer of 1946. Disposal of the fish was necessitated by lack of space. The data has not been analyzed as yet.

SECTION XII.

Chinook salmon adults, females and males, from Columbia River.

Treatment

15 fish were treated, four to 25 r, 7 to 50 r and 4 to 100 r, in September 1944 with 7 controls.

Spawning

September 21 to September 30, 1944.

Termination

Thirty-three fish were obtained from Spring Creek, a tributary to the Columbia River, and were hauled 270 miles by truck. This greatly reduced the vitality of the adults and their progeny. Thirty-three per cent of the mortality was due to transfer, hauling and accident. The other 67 per cent died naturally, during or after spawning. For further information refer to Section XIII.

SECTION XIII.

Chinook salmon eggs from Section XII. A total of 54,347 eggs was produced.

Spawning to eyed stage, September 21 to October 12, 1944; eyed stage to hatching, October 5, to November 9, 1944; hatching to feeding, November 9, 1944 to December 12, 1944; feeding to liberation, December 12, 1944 to June 29, 1945.

Termination

Mortality, growth and weight data were collected. Controls as well as irradiated groups suffered high mortality, so that deaths may be attributed to trauma rather than to irradiation. The fish were liberated June 29, 1945 in May Creek, a tributary to the Skykomish River.

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SECTION XIV.
Chinook salmon fingerling, female and male, (1/2 of Section XIII).

Treatment
Exposures of 25, 50 and 100 r were administered.

Termination
This section was cancelled due to lack of usable marks for migration studies.

SECTION XV.
Steelhead trout adults, females and males, from Columbia River (Leavenworth)

Treatment
16 fish were treated, four each to 25 and 50 r and eight to 100 r, in May 1944 with eight controls.

Spawning
Fish were spawned May 15 to June 2, 1944 and died naturally between May 14 and June 5, 1944.

Termination
The progeny are the subject of Section XVI.

SECTION XVI.
Steelhead trout eggs from Section XV. A total of 50,107 eggs were obtained.

Spawning to eyed stage, May 15 to June 14, 1944; eyed stage to hatching, May 30 to June 22; hatching to feeding, June 2 to June 28; feeding to liberation, June 28 to May 10, 1945.

Marking for Identification
3,057 of the 100 r group were marked by the removal of the adipose and left ventral fins. 3,285 of the control group were marked by the removal of the adipose and right ventral fins. The 25 and 50 r group lots were not marked before releasing.

Liberation
13,360 unmarked fish were planted in the Leavenworth Hatchery September 1944. These fish fish were from the 25 r and 50 r exposured parents and surplus controls. The marked fish were transferred to Leavenworth and liberated in the No. 3 holding pond in Icicle Creek.

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Returns

Six fish were recovered in the commercial fishery of the Columbia River during the fall of 1946, one in Russian River, California in the summer of 1947 and 12 fish were recovered in fall of 1947 in the Columbia River. Spawning takes place in the Columbia River in the spring of 1948.

Termination.

An attempt will be made to follow the development of any spawn procured.

SECTION XVII.

Steelhead trout fingerling, female and male, from Columbia River.

Treatment

Fish were exposed to 25, 50 and 100 r.

Termination

This section was cancelled due to lack of usable marks.

SECTION XVIII

Goldfish, females and males.

Treatment

Goldfish were exposed to 25, 50, 100, 250, 500, 750, 1,000, 1,250, 2,500 and control (size 1½") and 1,000, 1,250, 2,500 r and control (size 3"-4").

These fish were treated in December 1944 with 75 in each lot of the small groups except the 1,250 r lot which had 67 and 2,500 r lot which had 66. The larger fish had 21 fish in the 1,000 r lot, 19 fish in the 1,250 r group and 20 each in the 2,500 r lot and the control lot.

1,100 fish were purchased but many were lost due to parasites before treatment. The fish were received in October 144 but were not treated until December 1944 by which time the parasitic infection had been brought under control. Tissue was removed for sectioning (weekly). Mortality curves and kodachrome studies were made. The curves show the mortality to 105 days when $CuSO_4$ was inadvertently added to the City water supply.

Termination

Fish died December 1944 to May 1945 at which time the experiment terminated. The data accumulated should be summarized in a report and published.

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PROJECT II. Basic Studies of the Effect of X-Rays upon Planktonic Forms.

These experiments were undertaken to provide information on the sensitivity of marine micro-plankton to single mass dosages of X-rays.

Cultures of four organisms, Chlorella sp., Nitzschia closterium, a carteriid, and another mastigophoran (unidentified, brown), were subjected to exposures of 1,000 r, 10,000 r, 25,000 r, 50,000 r, or 100,000 r of X-ray. Two similar cultures of each organism were used as controls.

Numbers of organisms per unit volume of culture were counted using ruled blood counting chambers. Immediate immobilization of the brown mastigophoran was 50% effective at 50,000 r, and 100% at 100,000 r. Counts for an organism were comparable only with other counts for different irradiation samples of the same organism on the same date. Counts over a period of 4 months were most nearly as might be expected in the case of Chlorella where the concentration of individuals remained inversely proportional to the amount of irradiation for practically the entire time, except in the case of the 1000 r sample which exceeded one of the two controls. Nitzschia counts in the higher irradiations decreased earlier and more drastically than did those of Chlorella, but later these more heavily irradiated samples gave higher counts than the lower dosages. Counts of the two Mastigophora were arranged erratically with only a general tendency for count to be inversely related to exposure. In no case did a sample count drop to and remain at zero.

The results of this initial study were summarized into report UWFL-5, "Lethal Effect of X-Rays upon Four Marine Micro-Plankton Organisms".

The data were further reduced and published in Science.

Bonham, Kelshaw, Allyn H. Seymour, Lauren R. Donaldson, and Arthur D. Welander.
1947
Lethal effect of X-rays on Four Marine Micro-Plankton Organisms
Science Vol. 106, No. 2750, September 12, 1947

PROJECT III. Bikini Experiments of 1946.

Members of the Applied Fisheries Laboratory were present during the two Atomic Bomb tests at Bikini Atoll during the summer of 1946. During the test they joined the staff of the Division of Radiobiology of the Radiological Safety Section of Operations Crossroads.

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A preliminary report of the observations and findings was prepared and submitted with the reports of the Radiological Safety Section.

A vast amount of material was collected and remains to be worked up. This includes an extensive fish collection, tissues from animals before and after the tests, radioautographs, etc.

PROJECT IV. Bikini Experiments of 1947.

The Applied Fisheries Laboratory operating under Contract No. W-28-094-eng-33 with the Atomic Energy Commission assumed a major role in the Bikini Scientific Resurvey of 1947.

Studies were directed toward the determination of: (1) The presence or absence of radiation in the various marine organisms; (2) the distribution of radioactive substances in the plants and animals from different geographical locations within and near the atoll; and (3) the activity of fission products in certain tissues and organs.

The data collected were summarized in a preliminary report; UWFL-7 "Radiobiological Resurvey of Bikini Atoll During the Summer of 1947."

Additional reports are in preparation on various phases of this project. A vast amount of material and data needs to be worked up to better understand the problems developed by this very complex experiment.

PROJECT V. Absorption and Food Chain Studies.

It was believed that laboratory controlled experiments might bring about a fuller understanding of the Bikini problems and their interrelationships.

A series of experiments using Bikini "mud" as a source of fission products is underway. This work using available marine forms and holding them in small saltwater aquaria will enable us to study the problems of absorption and concentration of active materials as they pass through the food chain of these animals.

PROJECT VI. Exposure of Marine Invertebrates to X-Rays.

Studies were started January 15 and 16, 1948 when six groups containing 20± amphipods each were exposed to X-rays. The six doses were 150 r, 900 r, 6,400 r, 16,000 r, 32,000 r and 82,000 r. Six similar lots were used as controls.

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Athe end of February all of the amphipods subjected to 82,000 r, 32,000 r, and 6,400 r had died and a single specimen of the original 20 in the 16,000 r group survived. Mortalities were low in the 150 r (5%) and 900 r (21%) groups. Mortalities of the control groups ranged from 5% to 52% with an average of 24%.

Newly hatched young were found in numbers in the control groups, but among the irradiated groups, only the lowest (150 r) group showed reproduction.

It is assumed from this preliminary experiment that the 100% lethal dose is less than 10,000 r, and that more than 900 r inhibits reproduction. Another experiment is planned to determine lethal and inhibitory dosages more exactly.

PROJECT VII. Training Program.

The extreme shortage of trained personnel in the field of radiobiology, especially aquatic radiobiology had made it necessary for us to institute a training program to develop personnel capable of carrying on the work.

Arrangements have been worked out between the University of Washington, Graduate School and the Applied Fisheries Laboratory, and in some special cases the Hanford Engineering Works, for "on the job" training of graduate students.

This program has already resulted in the awarding of one Doctor of Philosophy Degree. Three men are at present working for their Ph. D. and others are in the early training phase.

PROJECT VIII. Cooperative Project with the Hanford Engineering Works.

The studies in this field are designed to follow changes in the Columbia River as a result of the discharge of effluent from the plants into the river.

The installation of the laboratory at 100 F (146 Building) has made it possible to carry out direct experimentation with animals using the effluent water.

Fine cooperation has existed between the Atomic Energy Commission, General Electric Company, and the University of Washington, Applied Fisheries Laboratory in the exchange of materials and information.

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INDEX OF REPORTS PREPARED AT THE APPLIED FISHERIES LABORATORY, UNIVERSITY OF WASHINGTON, SEATTLE

Report No.	Title
UWFL-1	Equipment and Procedures used in the Study of the Effects of Irradiation of Fish with X-rays.
UWFL-2	Studies of the Effects of Roentgen Rays on the Growth and Development of the Embryos and Larvae of the Chinook Salmon (Oncorhynchus tschawytscha).
UWFL-3	Section 10 of Series of Experiments involving the Effect of X-ray on Fishes: Fingerling Chinook Salmon (Oncorhynchus tschawytscha Walbaum)
UWFL-3a	The Effect of X-ray on Mortality, Weight, Length, and Counts of Erythrocytes and Hematopoietic Cells in Fingerling Chinook Salmon, Oncorhynchus tschawytscha Walbaum.
UWFL-4	Histological Effect of X-ray on Adult Male Silver Salmon, Oncorhynchus kisutch Walbaum.
UWFL-5	Lethal Effect of X-Rays on Four Marine Micro-Plankton Organisms.
UWFL-5a	Lethal Effect of X-Rays on Four Marine Micro-Plankton Organisms. (See page 15, manuscript was published)
UWFL-6	Section I and II of series of Experiments involving the Effect of X-Ray on Fishes: Chinook Salmon (Oncorhynchus tschawytscha Walbaum). Preliminary Report concerning X-ray Effects upon Chinook Salmon (Oncorhynchus tschawytscha Walbaum) observed through more than one Generation.
UWFL-7	Radiobiological Resurvey of Bikini Atoll during the Summer of 1947.
UWFL-8	The Effects of Roentgen rays on the Embryos and Larvae of the Chinook Salmon.
UWFL-9	Project Chronology Chart and Summary of the Applied Fisheries Laboratory, University of Washington, Seattle, Washington. Revised March 11, 1948

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DOE ARCHIVES

PUBLISHED REPORTS OF THE APPLIED FISHERIES LABORATORY, UNIVERSITY OF WASHINGTON, SEATTLE

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