

(13)

Original: Fish Division

cc: Education-Game

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Institute for Fisheries Research

INSTITUTE FOR FISHERIES RESEARCH

DIVISION OF FISHERIES

MICHIGAN DEPARTMENT OF CONSERVATION

COOPERATING WITH THE

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February 1, 1945

REPORT NO. 996

INVESTIGATION AND TREATMENT OF DISEASED BROOK TROUT

AT THE WOLF LAKE HATCHERY

by

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During the forepart of March 1944, Mr. Jay Marks, Regional Fisheries Supervisor at the Wolf Lake Hatchery, called the Institute's attention to a high mortality occurring among a stock of legal-sized brook trout being held in spring pond No. 3 at the Wolf Lake Hatchery. The writer arrived at the hatchery on March 8, 1944 and conducted an investigation of this mortality. Preliminary treatment, after diagnosis, was instigated on the same date with small samples of infected fish. On March 14 and 21 final treatment was administered. The results of the complete investigation are presented below.

According to Mr. Jay Marks about 35,000 legal-sized brook trout were being held in pond 3 over winter as 1944 planting stock. Hatchery attendants began noting an occasional dead trout in early February and reported a steady increase in mortality as the month progressed. Presented below is a daily record supplied by the hatchery of dead trout removed from pond 3 beginning on February 20 and extending through March 9.

Date	Air temperature (°F.)			Number of dead trout
	Morning	Noon	6:00 p.m.	
February 20	15	28	32	2
21	21	39	46	4
22	35	38	39	5
23	32	36	41	10
24	23	36	65	15
25	32	38	37	11
26	48	61	64	7
27	34	34	32	5
28	30	32	30	13
29	26	27	32	... *
March 1	20	28	54	48 **
2	27	36	40	9
3	38	40	40	7
4	30	30	28	14
5	1	21	34	15
6	30	35	38	60
7	20	22	20	36
8	17	20	18	17 ***
9	17	24	20	15 ***

* Were not collected that day

** Includes dead of both days

*** Includes dead and badly infected fish removed

Examination of pond 3 on March 8 revealed several dead trout present along the shores and lying on the bottom in shallow water. This pond covers an area of 150 by 50 feet with a maximum depth of 4 feet. The pond is supplied by spring water entering at a rate estimated at 500 gallons per minute. Most of the trout being held in the pond were congested in the upper third portion, that area nearest to the spring water inlet. Close observation of these fish showed that an estimated 50 to 75 per cent bore body disfigurements characterized by light colored blotches on the body and eroded fins. Many of these fish were seen to scrub themselves against the gravel bottom. By the use of a long handled dip net several of the more severely infected fish were collected and later examined under a microscope at the hatchery. In a discussion with Mr. Marks it was inferred that the water from pond 3 flows directly into a pond below containing yearling rainbow trout. Some of these fish were examined and

were found to be healthy in all respects, indicating that the epidemic was localized in the brook trout pond.

Results of microscopical examination of the diseased trout secured from pond 3 were as follows:

External condition

General: The dorsal and pectoral fins in all cases showed some erosion partially covered with a fungus growth and mucous secretion. Several irregular bluish-grey patches were noted on the body of each fish, usually located near the base of the dorsal or midway between the dorsal and pectoral fins. Areas of the body not involved in the infection bore a natural color. The fins also were of a healthy red color and no signs of infection were present.

Microscopical: Infected appendages were examined and found to harbor Gyrodactylus.^{*} These flukes were quite numerous; sometimes a total of 40 individuals were counted on one fin. In addition to this parasite, there were heavy growths of fungus and a stalked protozoan (Epistylis)^{*}, located about the infected area. It is possible that the latter two organisms are secondary infections. Examination of the irregular skin blotches revealed the presence of the same organisms as were described on the fins.

Internal examination

General: The stomach appeared normal, partially digested food being present. The intestines were covered with mesenteric fat and were of normal appearance with the exception of a small portion near the posterior end which looked highly inflamed. The liver and kidneys were of a normal brownish-red color and presented no indication of being infected.

^{*} Specimens of the living parasites were brought to Ann Arbor and examined by Dr. George R. LaRue, Professor of Zoology at the University of Michigan, who verified the identity.

Microscopical: A portion of the hind gut extending from the vent forward for two or three inches, mentioned as highly inflamed in the above paragraph, was found to contain several spiny-headed worms. These worms are armed with several rows of hooks which were found to be deeply embedded in the intestinal mucosa. This highly inflamed condition of the intestine could have been caused by this organism.

In control of the ecto-parasites, three methods of treatment were tried on small batches of infected trout. A total of 36 infected fish were collected and divided into three equal groups and treated in the following manner: Group 1 was immersed in a formalin bath for 60 minutes at a concentration of 1 to 4000. Group 2 was immersed for one minute in a solution of glacial acetic acid (1 to 500), one part of acetic acid to 500 parts of water. Group 3 was placed in a saturated solution of sodium chloride for a period of five minutes. The next day (March 8) these fish were examined and two of the above treatments were found to be unsatisfactory. Group 1, treated in the formalin, had a loss of 6 fish, but all of the ecto-parasites were killed. Group 2 (treated in acetic acid) had a loss of 2 fish, and 4 of the remaining 10 fish still harbored living parasites. No loss of fish in Group 3 was encountered nor was there any appreciable reduction in the number of ecto-parasites on the body. This method of treatment apparently has no therapeutic value as far as control over external parasites of this type.

In as much as the formalin treatment was the most effective in ridding the fish of these parasites, further studies were in order as a reduction in the initial deaths was desirable. It was thought that perhaps if a shorter exposure period during treatment could be worked out satisfactorily, a reduction in the initial mortality might be achieved.

A total of 7 infected trout were secured and marked individually for later identification and given a 1 to 4000 formalin treatment ranging individually for a period of 20 to 50 minutes. These fish were treated on March 8, 1944 and examined the next day. Presented below in tabular form are the results of this study.

Specimen number	Duration of bath exposure	Number of <u>Gyrodactylus</u> after treatment		Stalk protozoans
		Alive	Dead	
1	20	4	3	Present (alive)
2	30	1	0	
3	30	0	1	
4	40	0	7	
5	40	0	4	
6	50	0	12	Present (dead)
7	50	0	16	Present (dead)

Before treatment each of the above fish was examined and was known to harbor large numbers of Gyrodactylus. This was not the case after treatment, as evidenced by the above findings. It is probable that many of the flukes were effected by the chemical and became detached from the trout, thus accounting for the lower numbers. From the above data it appears that a treatment period of less than 40 minutes would be unsatisfactory. As no fish were lost in the 40 or 50 minute treatment and the parasite removal was complete, it was decided to use at least a 40 minute exposure when treating the total lot of trout.

Raceway number 6, the last one in a series immediately below the hatchery, was selected for the treatment pond. The water from this pond passed directly into the lake and did not enter into any of the other pond supply system. We felt that in using this pond for treatment the possibility of spreading the infection to other ponds would be greatly reduced. The total lot of trout (17,000) were removed from pond 3 and placed in no. 6 for treatment. The inlet and outlet of this pond was closed and the total volume was calculated which amounted to 42,185 gallons.

No additional water was allowed to enter or leave the pond during the treatment process with the exception of that used near the end of treatment (see below). By adding 10.5 gallons of formalin to the treatment pond, the desired strength (1 to 4000) was attained. The concentrated formalin was diluted (10 to 1) before being administered. The application consisted of carrying the diluted formalin in five gallon pails and emptying the contents with a swishing motion. It required $4\frac{1}{2}$ minutes to complete this process. In order to conform with a 40 to 50 minute treatment, lowering of the pond was started after a lapse of 33 minutes. The pond volume was reduced two-thirds in $7\frac{1}{2}$ minutes and fresh water being held back in race 4 (located immediately above race 6) was allowed to enter. It was estimated that the total operation required a period of less than one hour.

Samples of treated infected trout were examined the next day, March 15, and about 50 per cent of these fish were found to still harbor living Gyrodactylus. The initial loss of 50 trout was lower than expected and the ineffectiveness of the treatment was evident. It is possible that many of the infected trout were able to escape the formalin bath sufficiently long enough to prevent complete parasite removal. The complete mixing of the formalin and water was probably a much slower process than anticipated and our calculated volume of water in the pond may have been in error. In as much as the pond treatment was not a success, a trial tank treatment was proposed. As a trial run, 1600 trout were removed from the pond treated stock and placed in a hatchery unit containing a formalin solution of (1 to 4000) for a period of 40 minutes. These fish were then placed in race 4 for observation.

Mr. Marks and hatchery employees kept a record of the mortality for the two groups of treated fish in races 4 and 6 and furnished us the following information:

March 16 - Loss of trout in raceway	6 [*]	- 91
March 16 - " " " " "	4 ^{**}	- 15
March 17 - " " " " "	6	- 82
March 17 - " " " " "	4	- 0
March 18 - " " " " "	6	- 84
March 18 - " " " " "	4	- 0

✓* Pond treated on the 14th.

✓** Tank treated on the 15th.

In consideration of the above figures, the tank treatment appeared to be the more effective of the two methods. Though 15 trout were recorded as dead the next day from the original 1600, the mortality the next two days was zero. The 15 fish undoubtedly represent the initial loss following the treatment. Results of this trial tank treatment warranted an additional treatment for all of the remaining trout, which was undertaken on March 21 and completed on the 22d.

Two hatchery planting units each capable of holding 800 of these fish for one hour were employed. The tanks were filled to the desired height with a (1 to 4000) formalin solution and the fish introduced; after a 40 minute lapse they were removed and a new bunch added. By using the two units no time was lost during the treatment process. While one lot of fish was undergoing treatment, the other unit was being unloaded of its treated fish and a new stock collected. After two groups of fish had been run through each unit, the old formalin solution was drawn off and a fresh supply added. In this way the desired concentration of formalin was more or less constant.

After this treatment Mr. Marks kept a daily mortality record for the total stock of treated fish and furnished us with the following data:

March 22 - Loss of trout	36	March 27 - Loss of trout	9
March 23 - " " "	11	March 28 - " " "	15
March 24 - " " "	18	March 29 - " " "	17
March 25 - " " "	19	March 30 - " " "	3
March 26 - " " "	12	March 31 - " " "	1

Later examination of several of the trout which still bore marks of the previous infection failed to produce a single Gyrodactylus. By the end of March the death rate was very low and the infection appeared to be checked.

Summary

Yearling brook trout being held in pond 3 were found to be severely infected with Gyrodactylus. In addition, fungus and a stalked protozoan (Epistylis) were found on the same fish in many cases.

Control of these ecto-parasites was achieved by the use of a formalin bath (1 to 4,000) for a 40 minute immersion. Glacial acetic acid and brine baths were found to be ineffective in the control of Gyrodactylus, fungus and Epistylis.

Though pond treatment may be practicable under some conditions, it was found to be unsatisfactory on this occasion.

Whether the high mortality exhibited among the trout was due to the presence of Gyrodactylus was not determined. It is possible that the stalked protozoan may have had a deleterious effect producing toxic symptoms.

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